Supporting information

Naphthylthiazoles:Broad Spectrum Class of Antifungals

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Initial Antimicrobial Assessment and Establishing SAR. All synthesized compounds were tested against two Gram-positive bacterial strains (MRSA USA300 and *Clostridium difficile*), two Gram-negative bacterial strains (*E. coli* JW55031 (*tolC*-mutant) and *E. coli* BW25113 (wild-type)), and one fungal pathogen (*Candida albicans* SC5314) (Table 1). The lead ethylenediamine-containing compound **7** showed mixed modest activity against MRSA USA300 and *C. albicans* with an MIC value of 32 μg/mL (Table 1). To test whether the terminal amine was essential for the antimicrobial effect, this particular amino motif was removed and replaced with a methyl moiety (compounds **8** and **9**), and both compounds were void from any antimicrobial properties (Table 1). Therefore, we concluded that the two nitrogen centers of the lead compound's ethylenediamine side chain were essential pharmacophoric elements. Cyclization of ethylenediamine in the form of aminopyrrolidine remarkably enhanced the antifungal activity of compound **10** (MIC = 8 μg/mL, four times less than the lead structure **7**) and nullified the antibacterial activity against MRSA USA300 (MIC >64 μg/mL).

Next, we maintained the pyrrolidine scaffold and changed its substituents in which the free amino group was replaced with dimethylamine, amide or hydroxymethyl moieties (compounds **11**-**16**). In all cases, the distance between the pyrrolidine nitrogen and the terminal functionality (NMe_2, NH_2 or OH) was kept at two carbon units. So far, the amide-containing derivatives (compounds **13** and **14**) and their hydroxymethyl analogs (compounds **15** and **16**) were inactive. Only the *R*-dimethylaminopyrrolidine-containing derivative **12** revealed moderate antimycotic potency with an MIC value of 8 μg/mL. Notably, the *S*-isomer **11** was more selective towards MRSA with an MIC value of 4 μg/mL. These two observations further support our first assumption that the free rotation of ethylenediamine moiety of the lead compound **7** gives it the flexibility

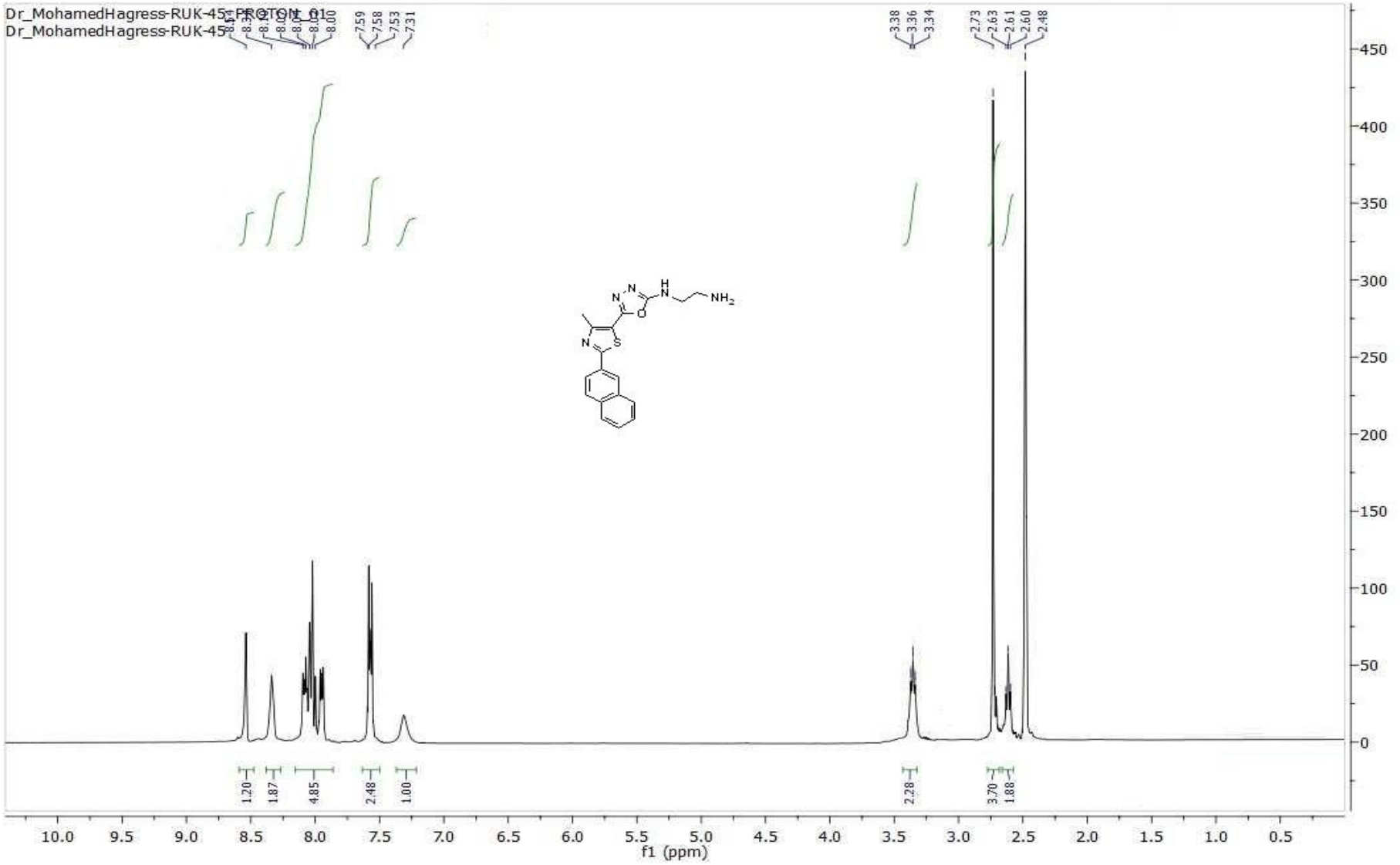
to have dual antibacterial/antifungal effects, and the moderate potency may be due to the high energy penalty required for transformation from the lower energy conformation (state in solution) to the bioactive shape.

With these promising results in hand, we expanded the size of the ring linker that connected the two amines together. Two diaminocyclohexyl moieties were tested with two different spatial configurations (compounds **17** and **18**). Interestingly, both compounds showed considerable improvement in antimycotic activity with MIC values against tested candida of 1 μg/mL. This value was 32-times more potent the lead compound **7** and similar to the drug of choice in this case (amphotericin-B). To test the effect of the distance between the two nitrogen centers carried on the cyclohexyl moiety, compound **20** with 1,4-diaminocyclohexane was prepared and its antimicrobial profile was tested. The MIC values of compounds **20** indicated that lengthening the distance between the two pharmacophoric amino groups significantly deteriorated the antimycotic effect (MIC = 64 μg/mL). Therefore, the essential two-carbon distance was fixed and an additional more flexible derivative that contained aminomethylpiperidine (compound **19**) was also tested and was devoid of activity.

The last approach in our SAR study was to keep the two-carbons unit spacer and replace the terminal amine with an imidine motif. In this case, the two nitrogen atoms of the imidine moiety covered a larger space. Thence, the imidine **21** showed no improvement in the MIC value against the tested *Candida* strain compared with lead compound **7**.

Table 1. Initial screening (MICs, in μg/mL) of the newly synthesized naphthylthiazoles against bacterial and fungal strains

Tested compounds/ control drugs	**MRSA USA300**	***E. coli* JW55031**	***E. coli* BW25113**	***C. difficile* ATCC BAA1870**	***C. albicans* SS5314 (wild-type)**
7	32	64	>64	64	32
8	64	64	>64	64	>64
9	>64	>64	>64	>64	>64
10	>64	>64	>64	>64	8
11	4	32	>64	16	32
12	>64	>64	>64	>64	8
13	>64	>64	>64	>64	>64
14	>64	>64	>64	>64	>64
15	>64	>64	>64	>64	>64
16	>64	>64	>64	16	>64
17	>64	>64	>64	64	**1**
18	64	>64	>64	32	**1**
19	>64	>64	>64	32	>64
20	64	>64	>64	64	64
21	>64	>64	>64	64	32
Linezolid	1	8	>64	1	NT
Vancomycin	1	NT	NT	NT	NT
Gentamicin	NT	≤0.5	≤0.5	NT	NT
Fluconazole	NT	NT	NT	NT	0.5
Amphotericin B	NT	NT	NT	NT	1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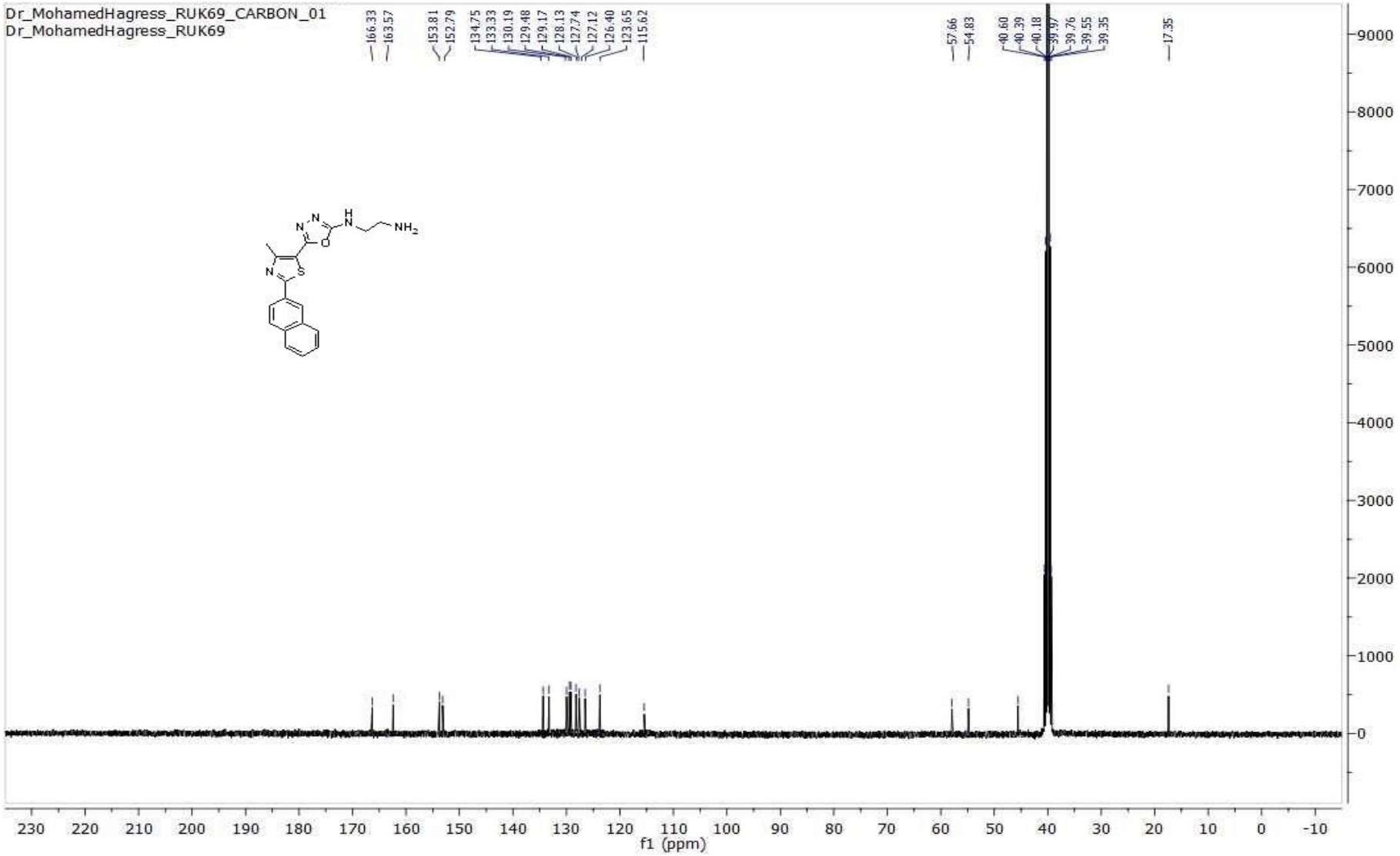
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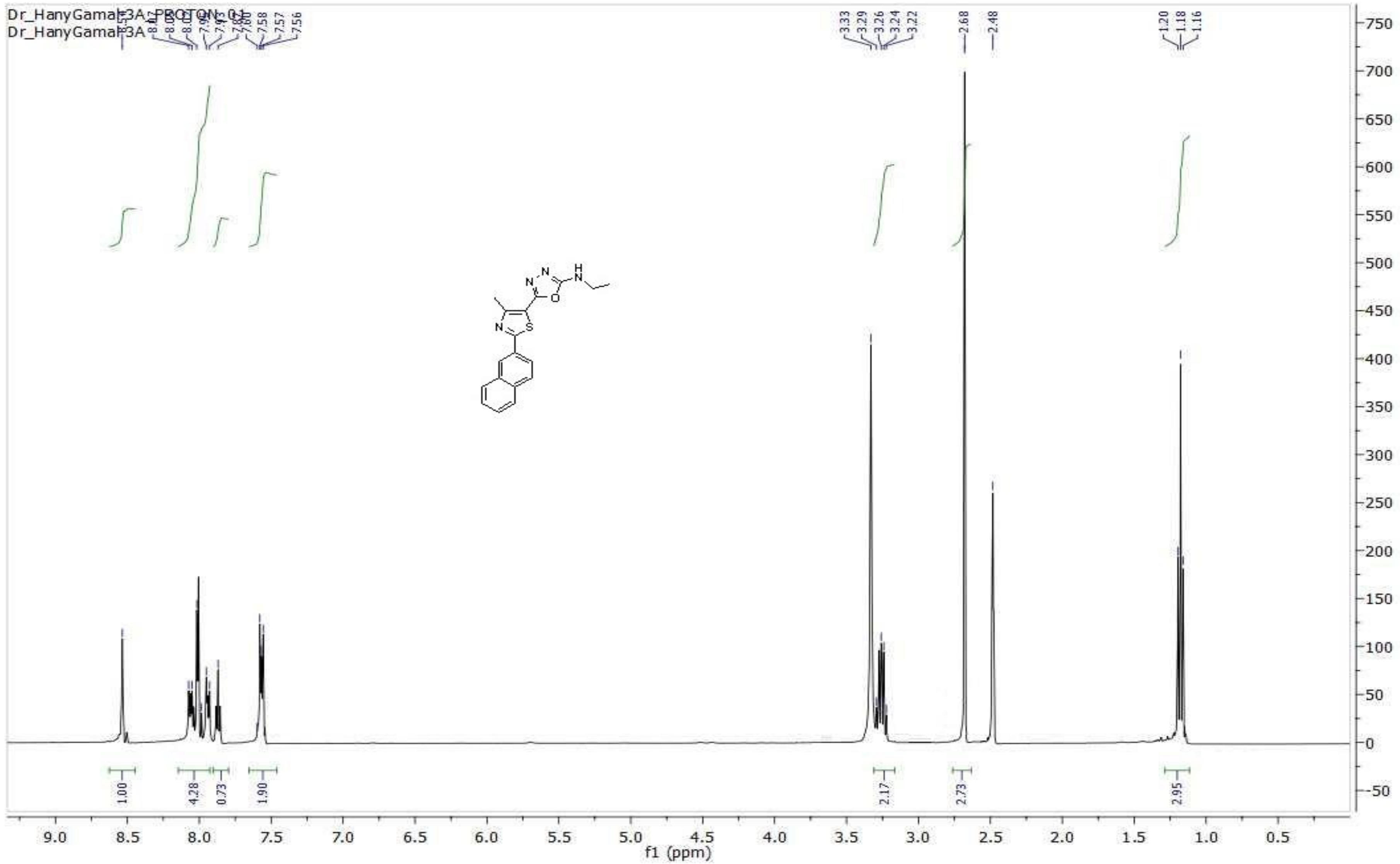
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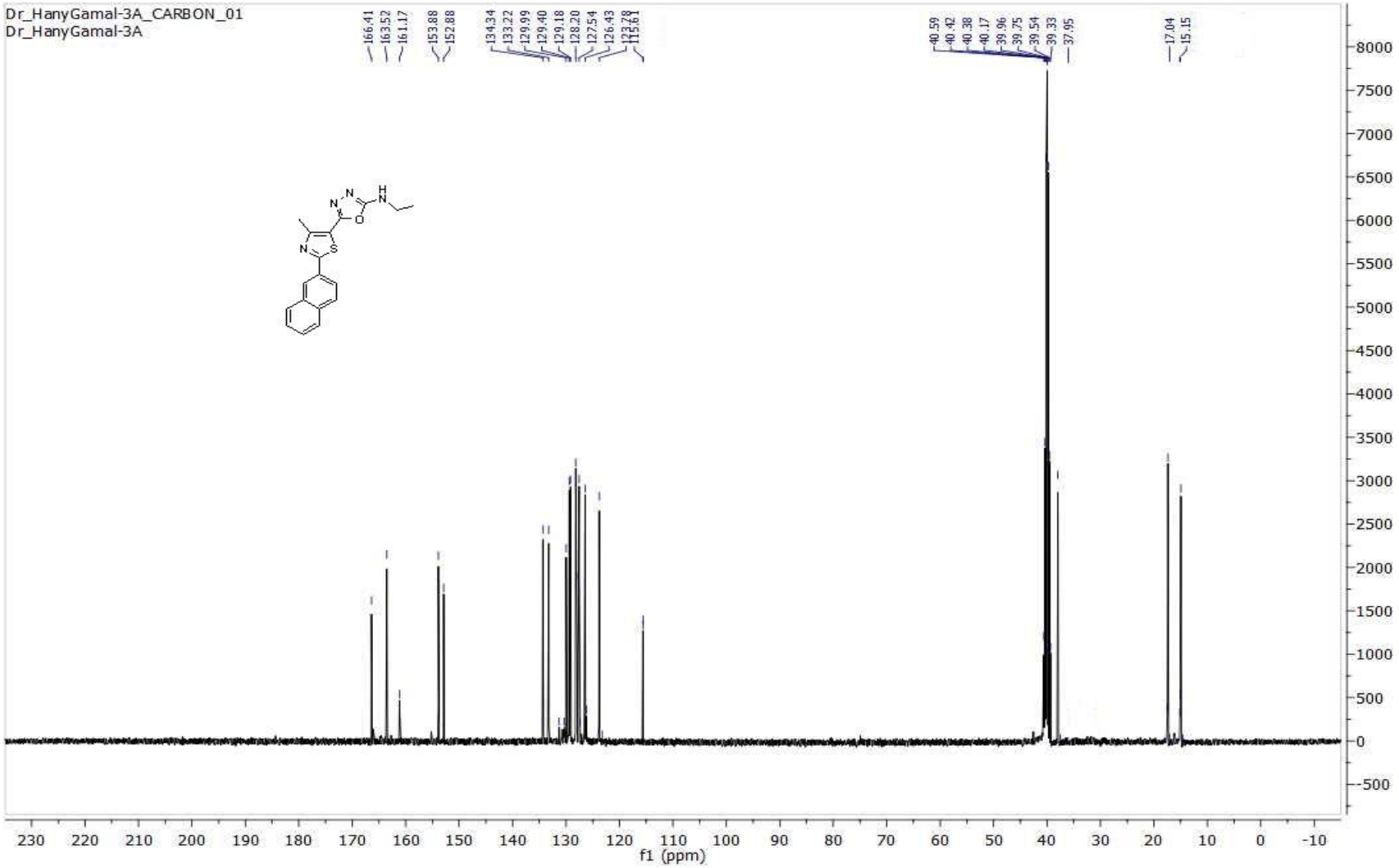
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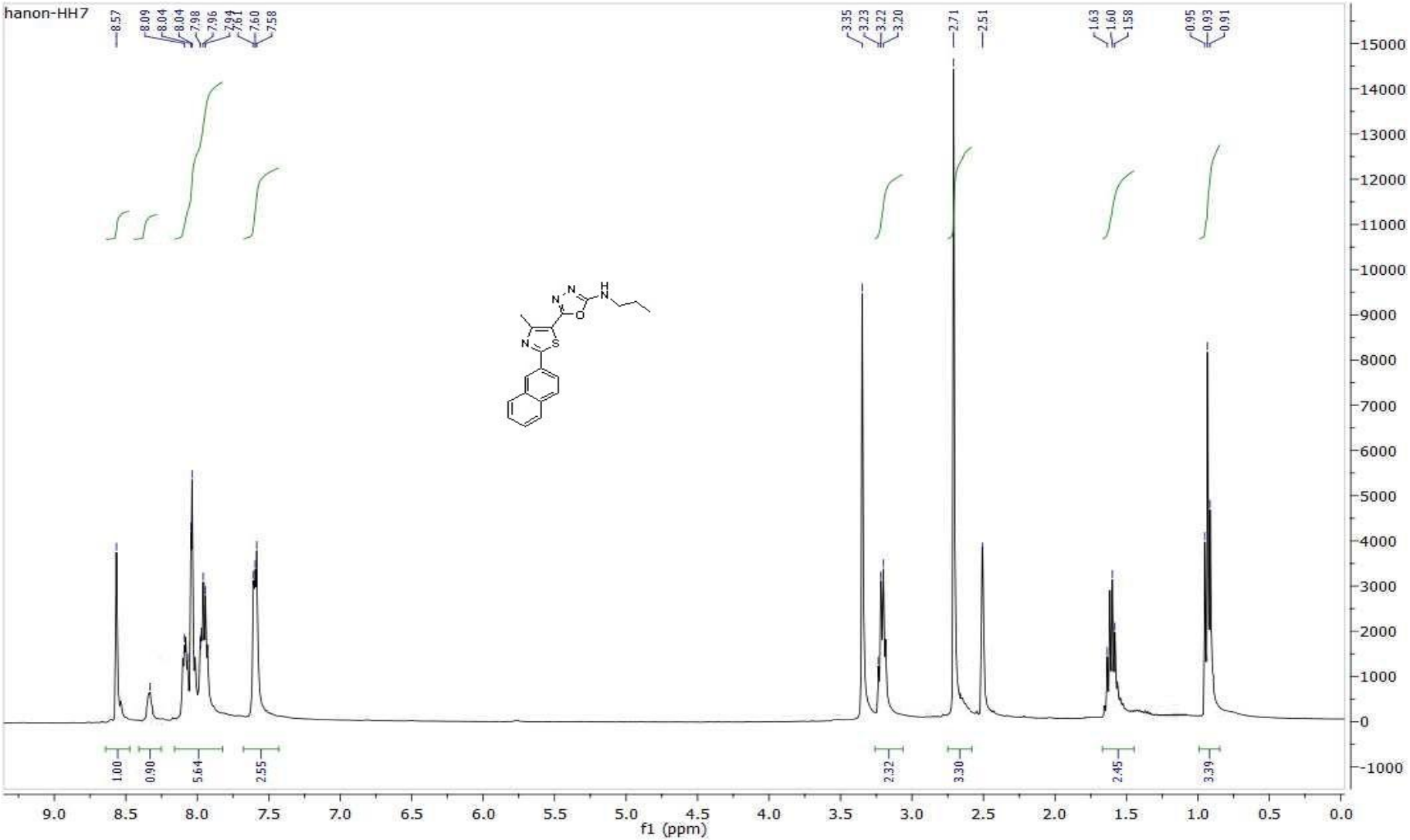
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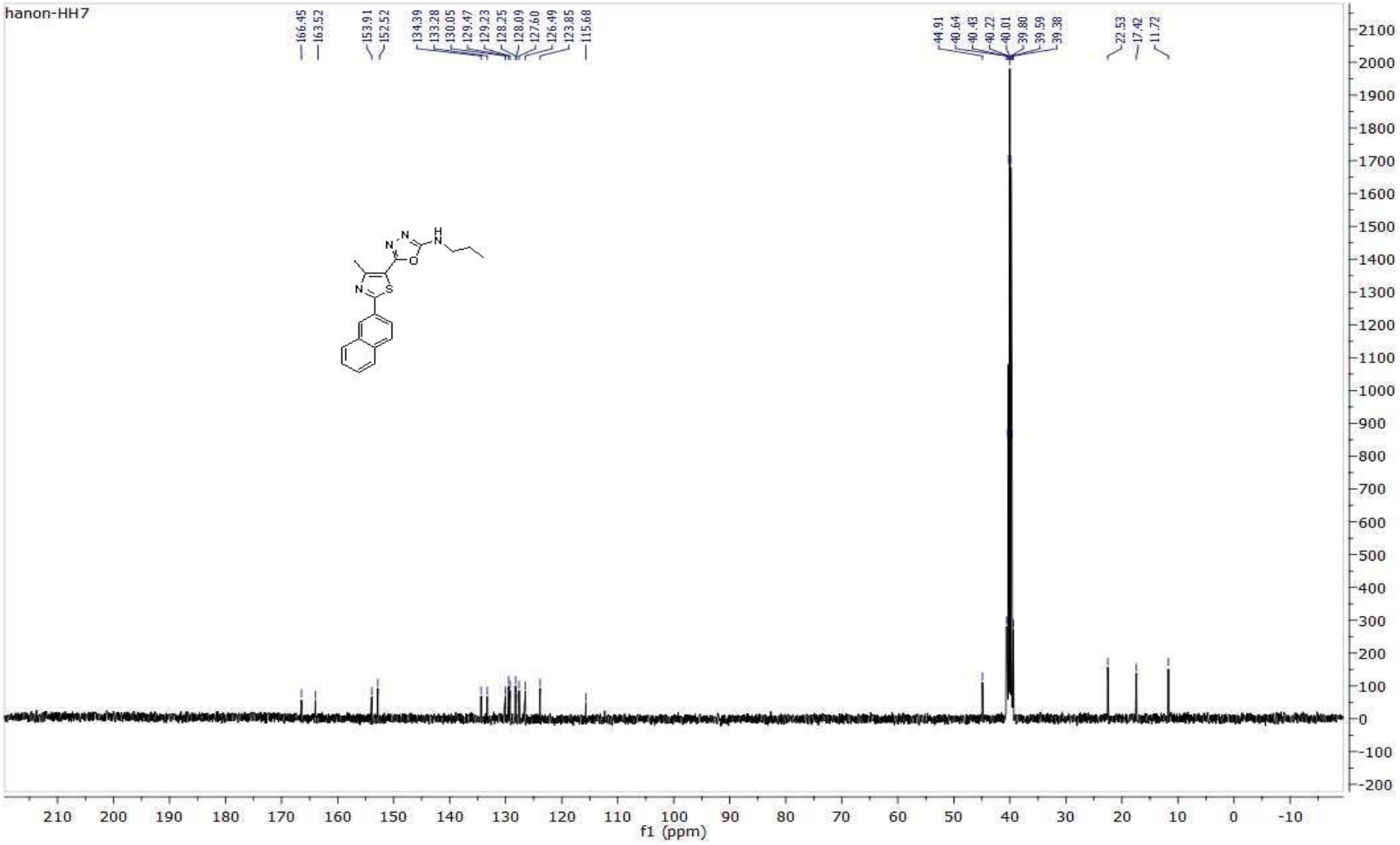
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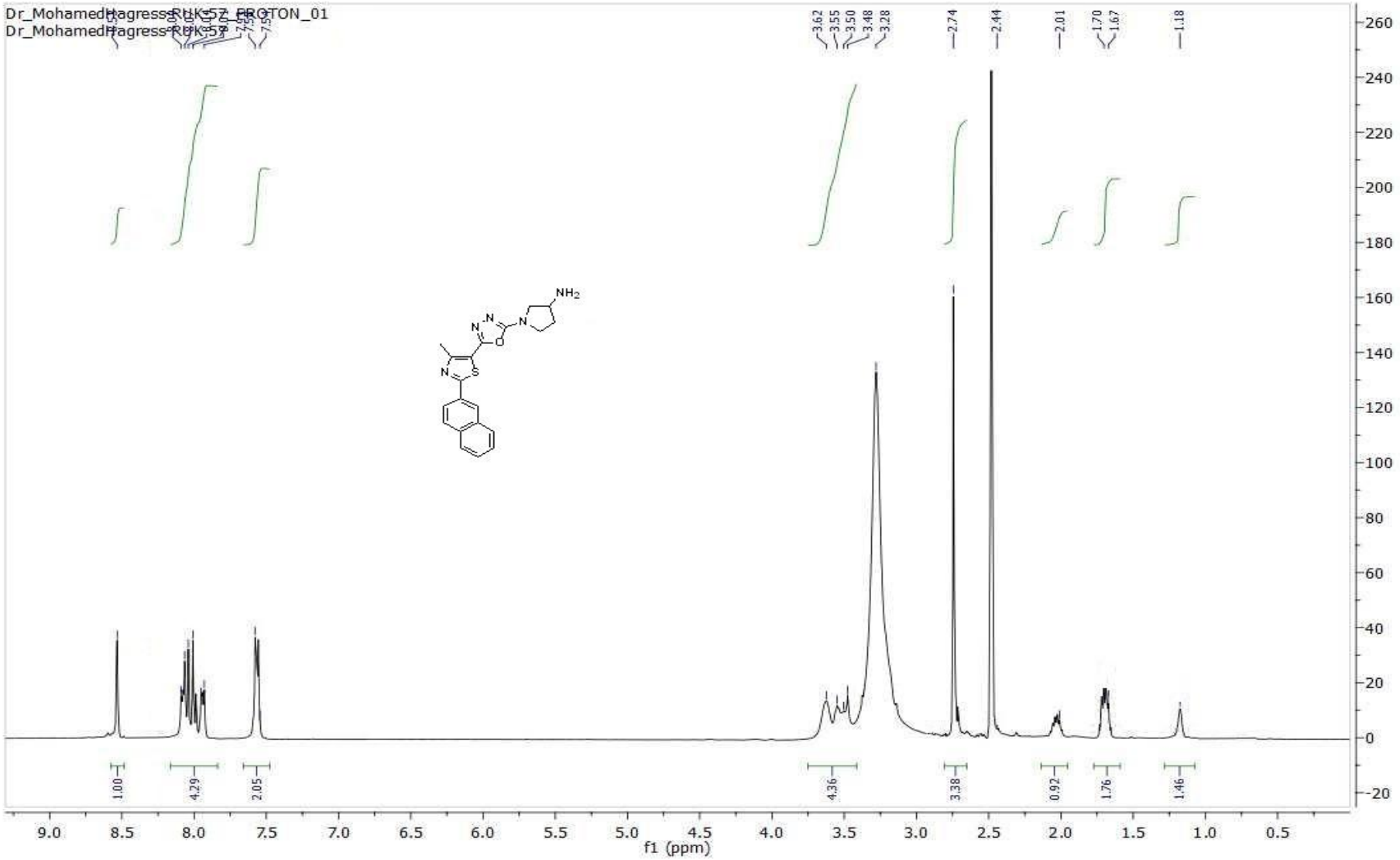
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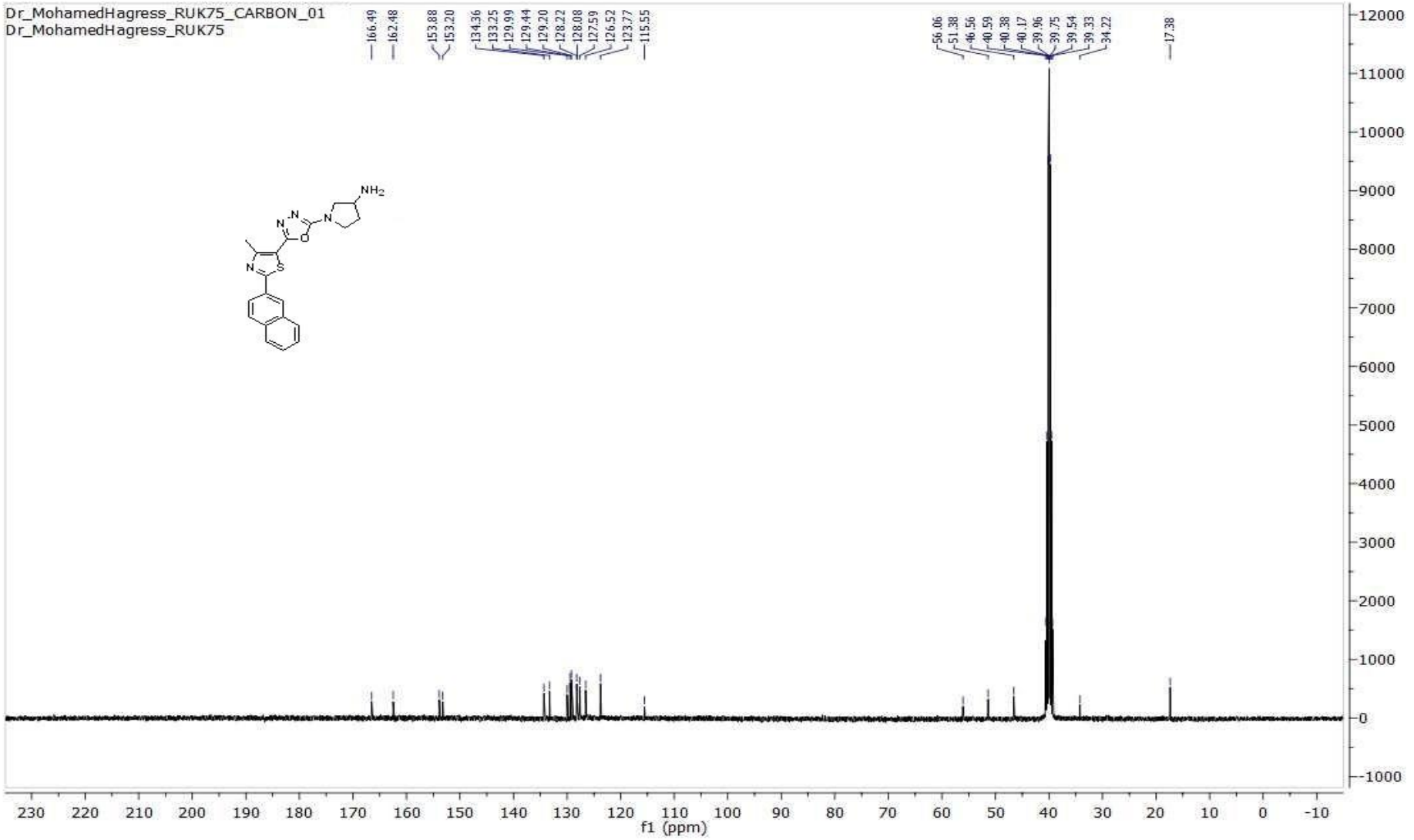
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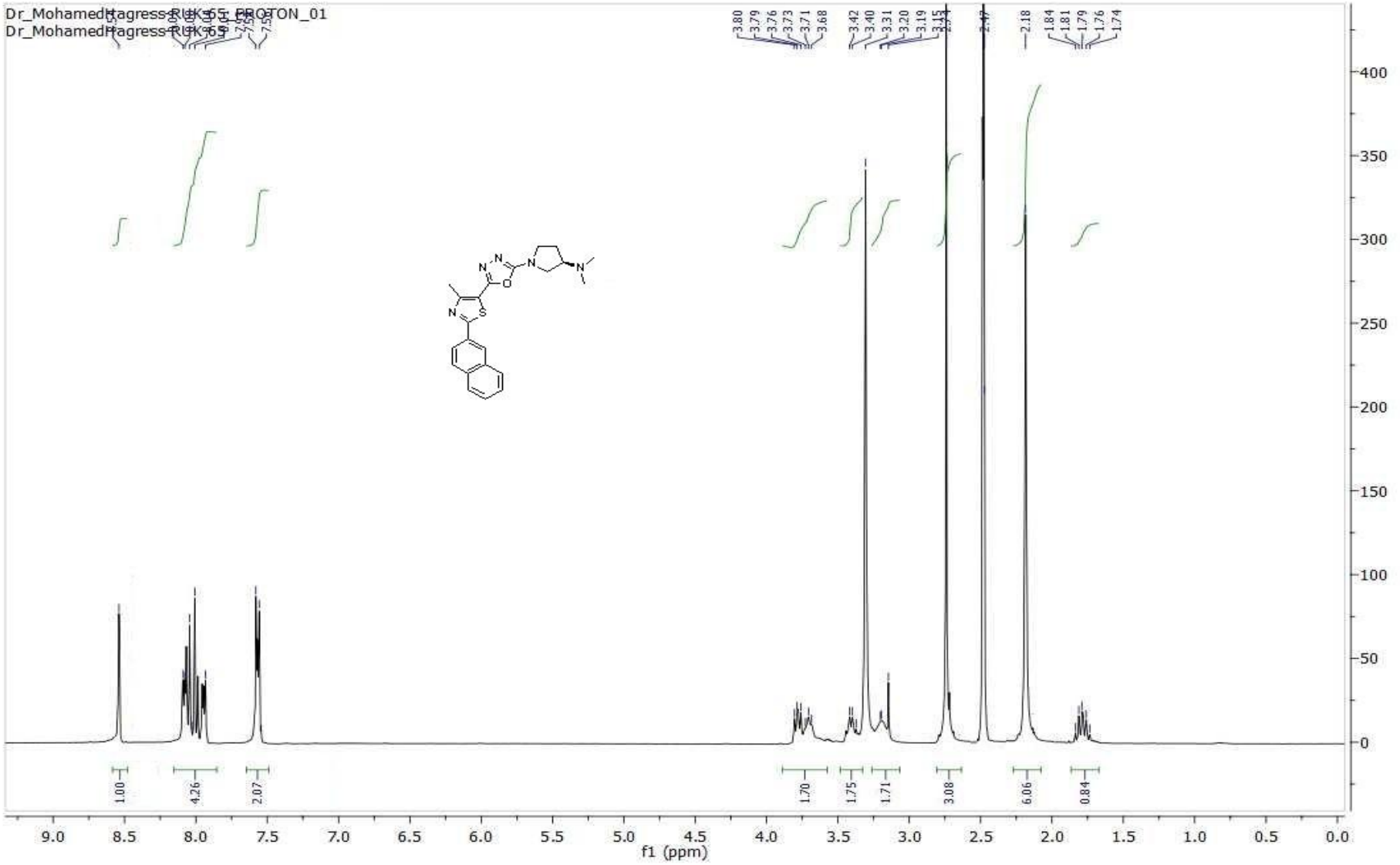
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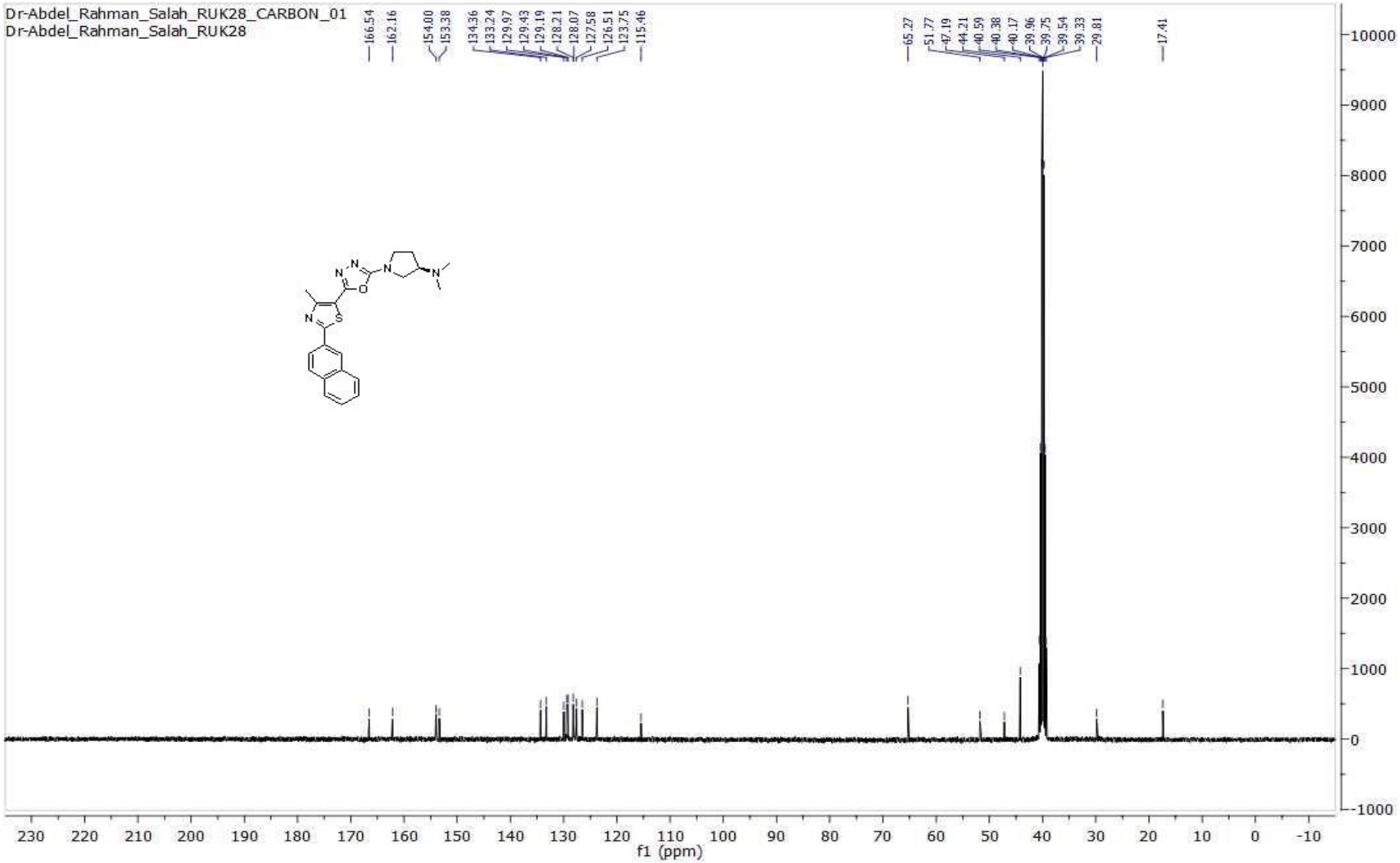
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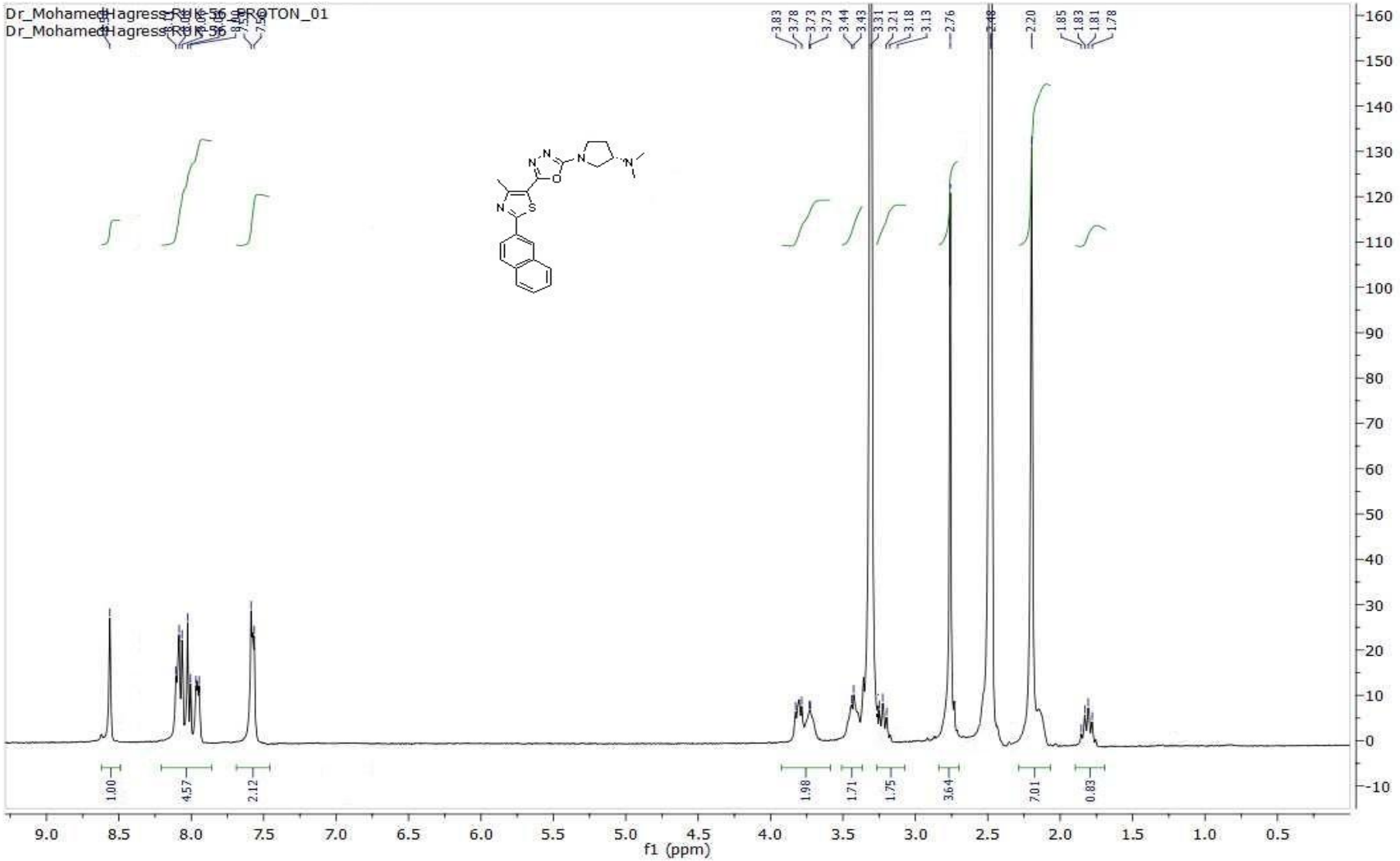
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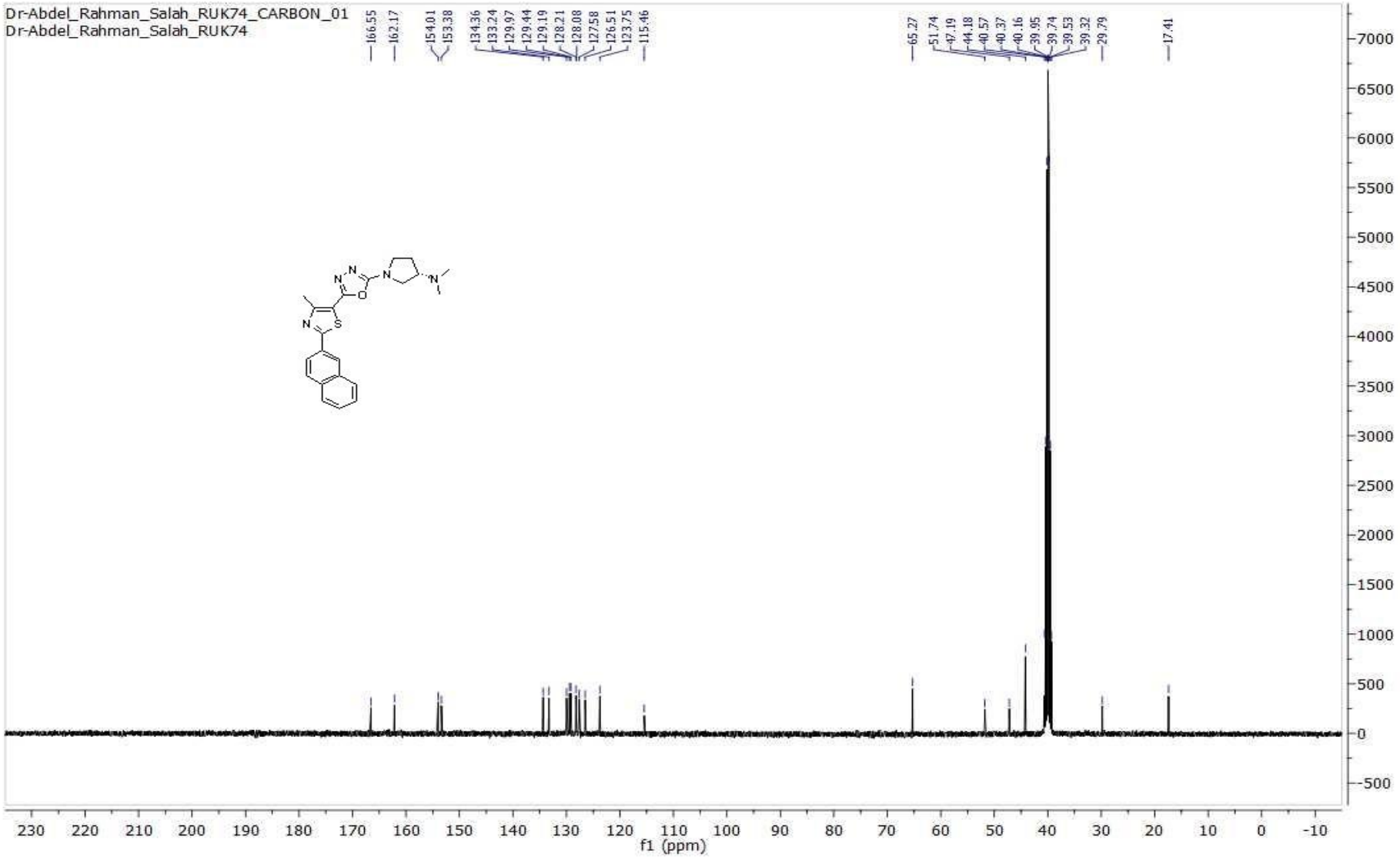
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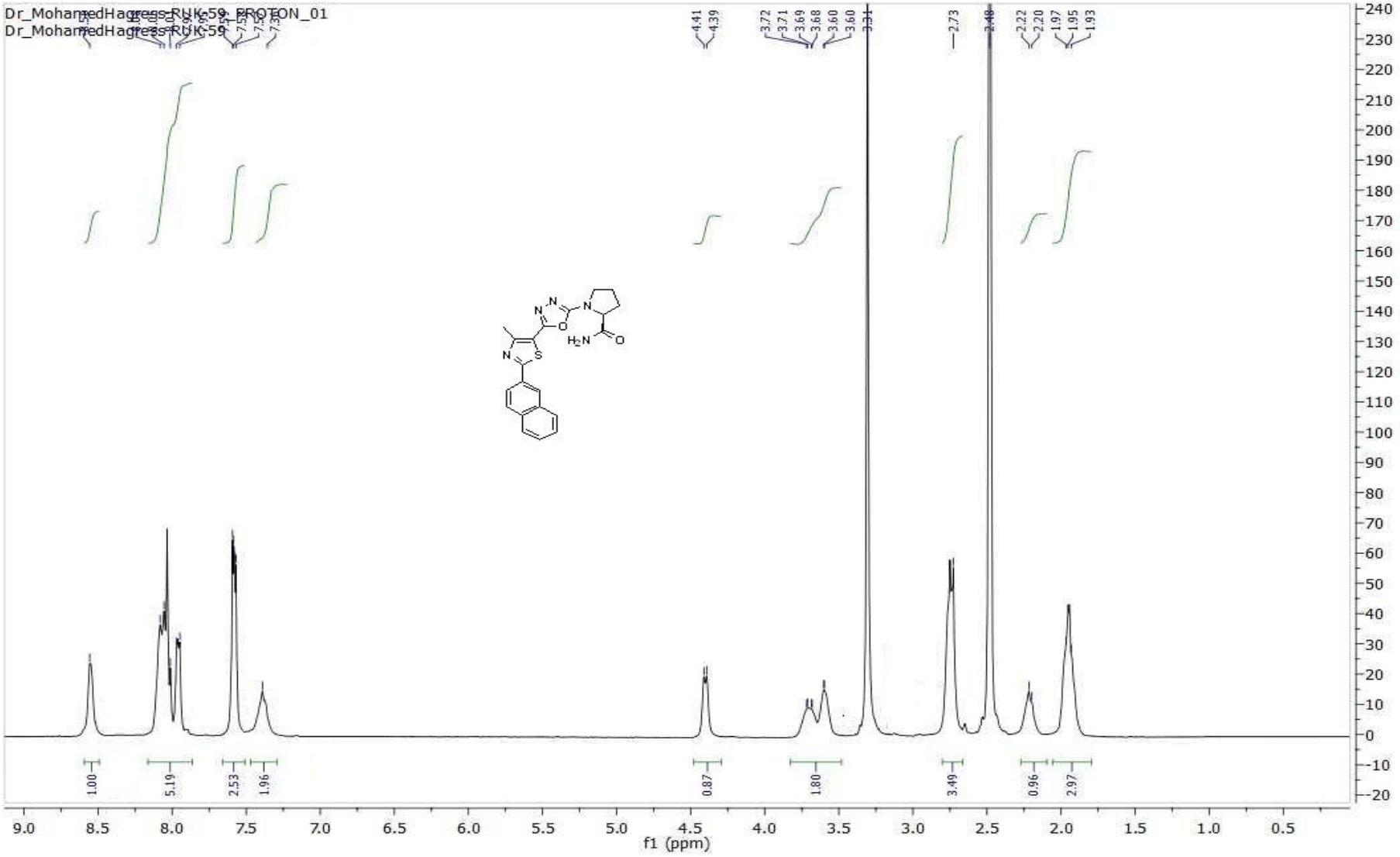
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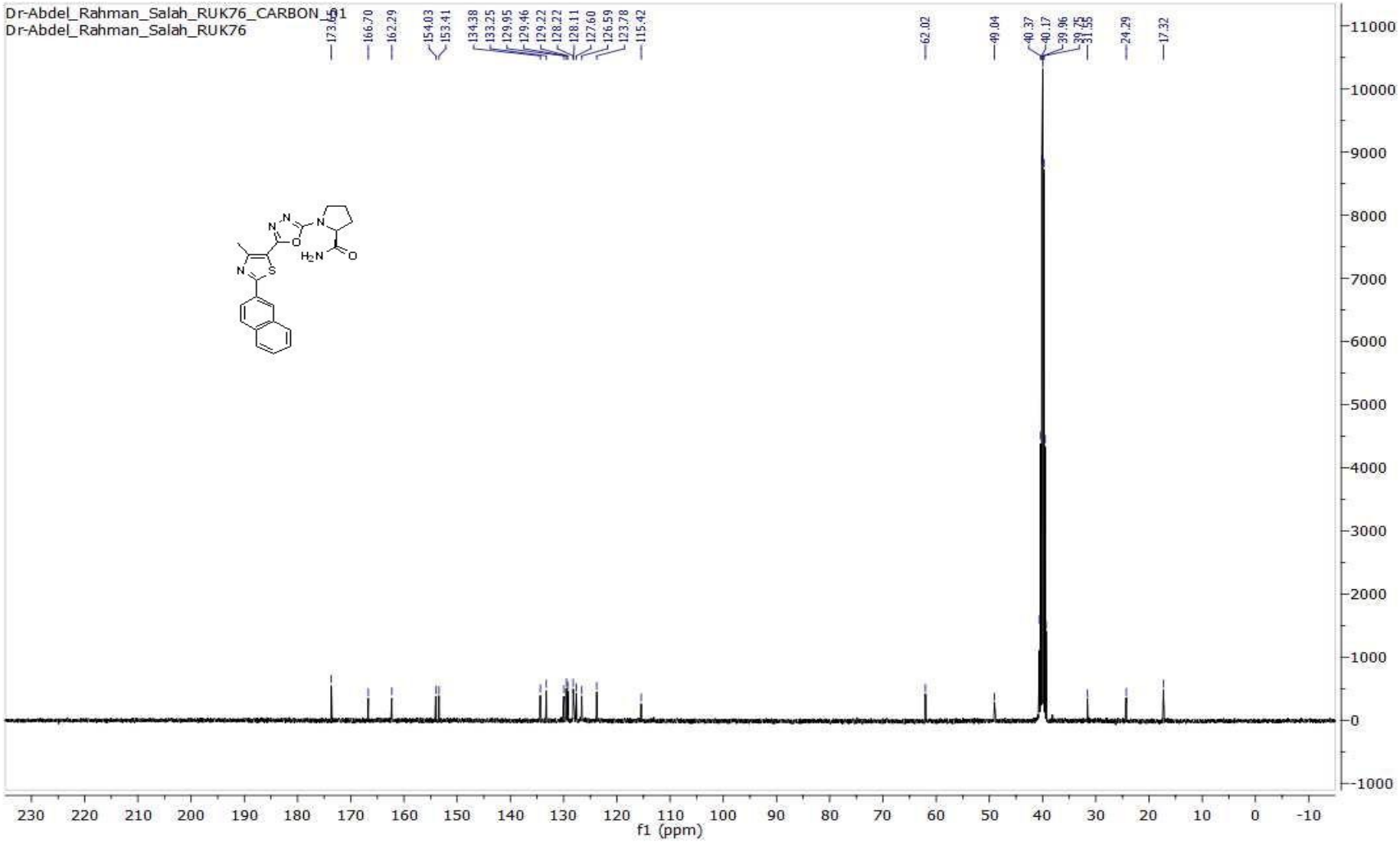
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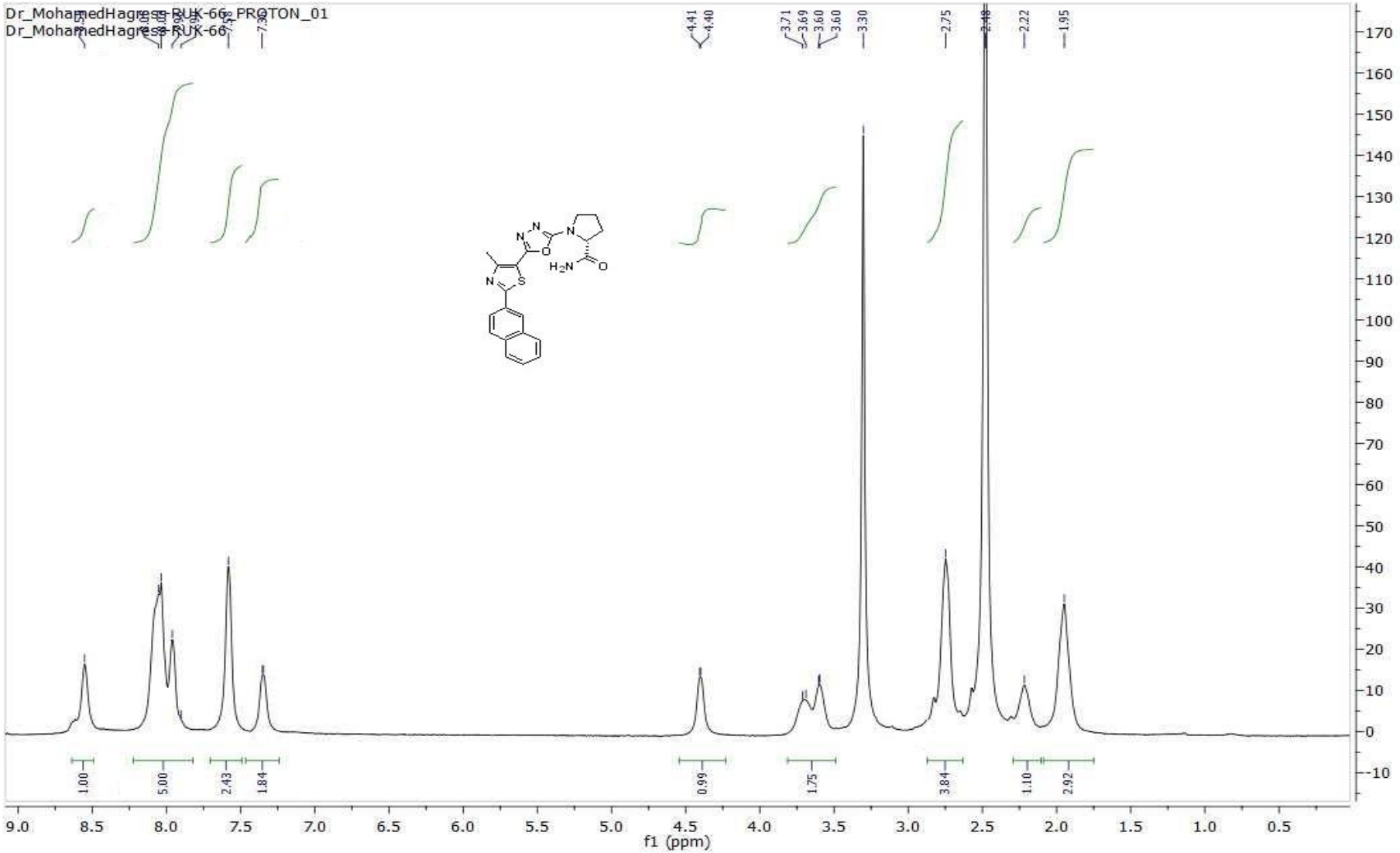
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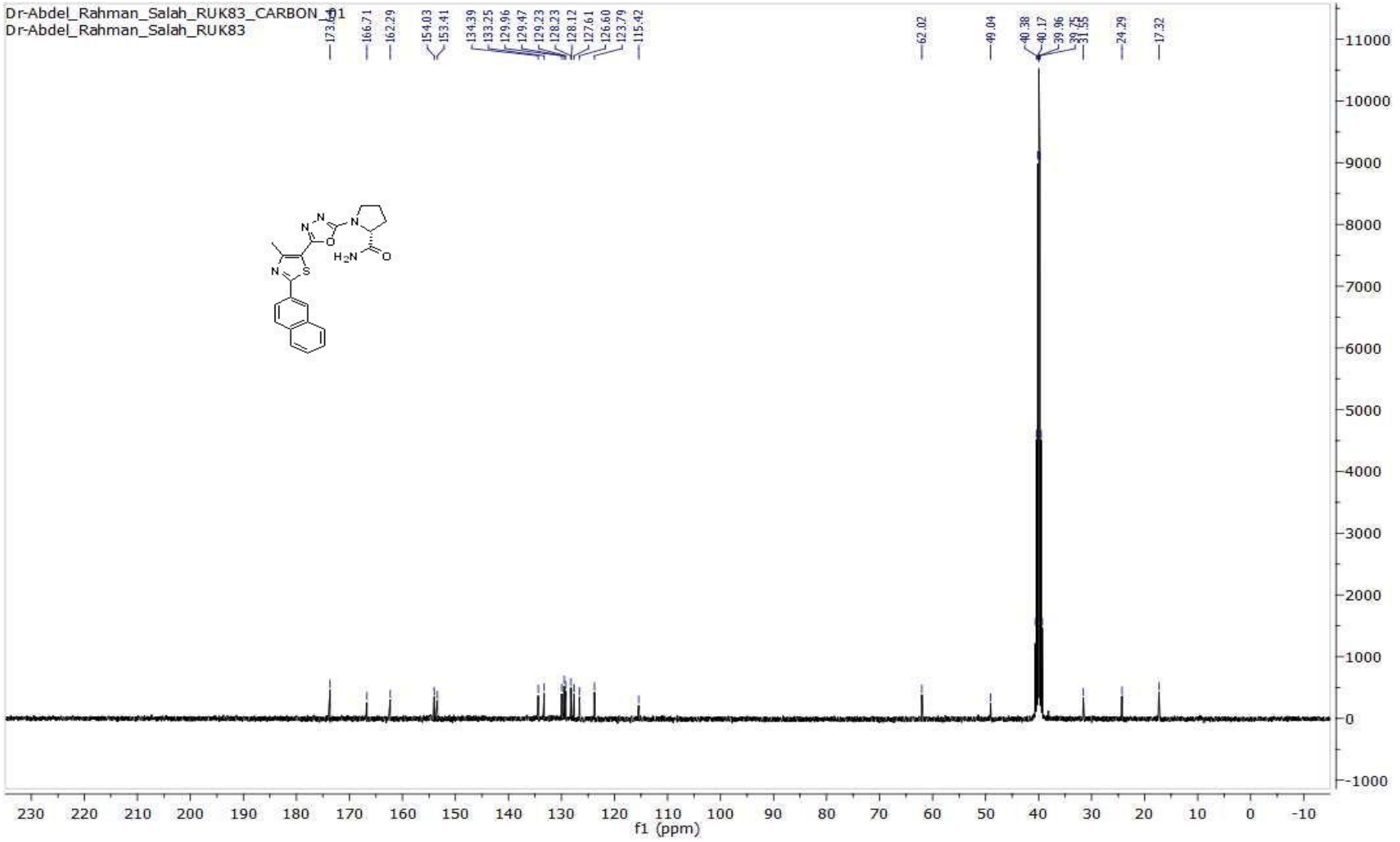
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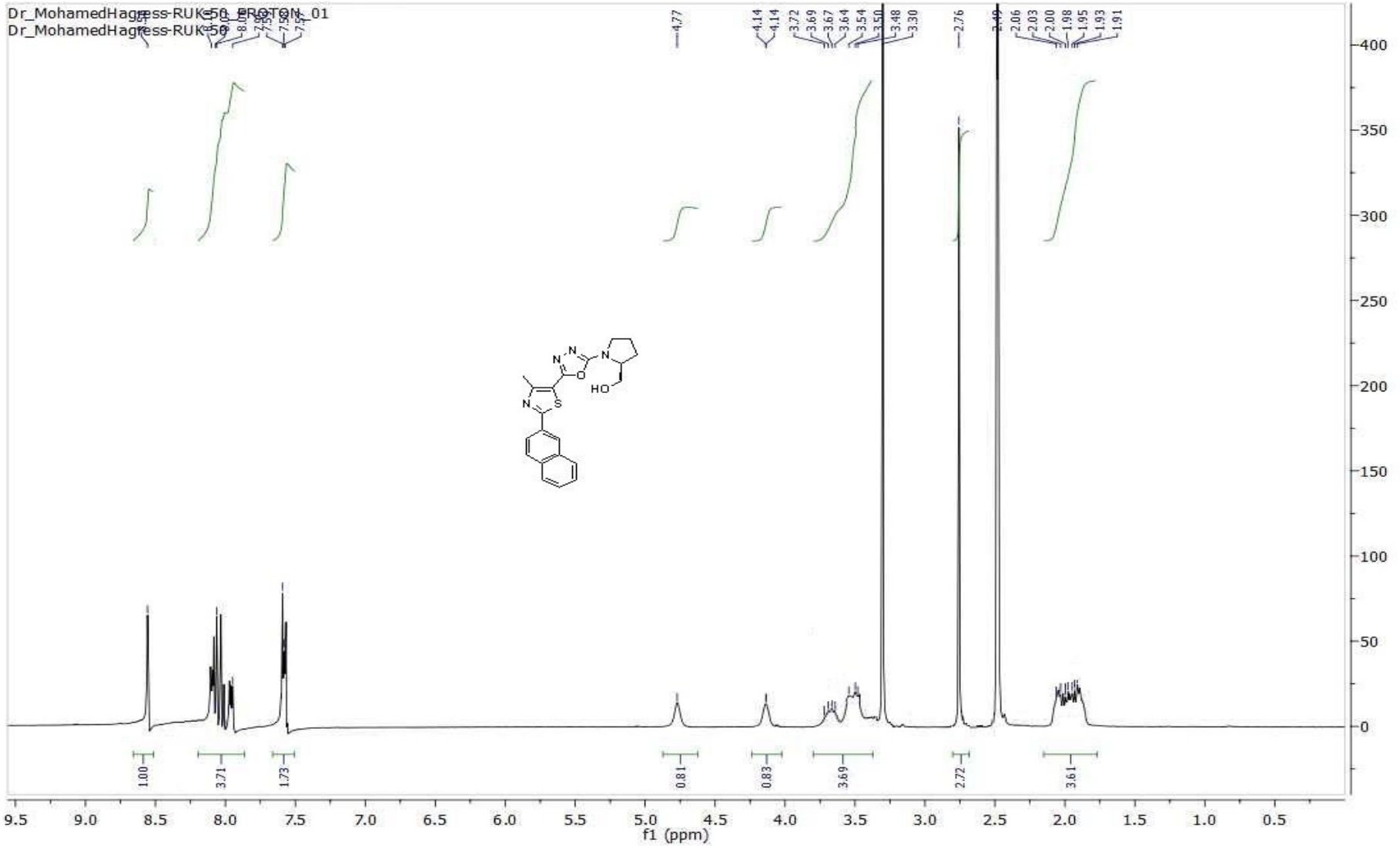
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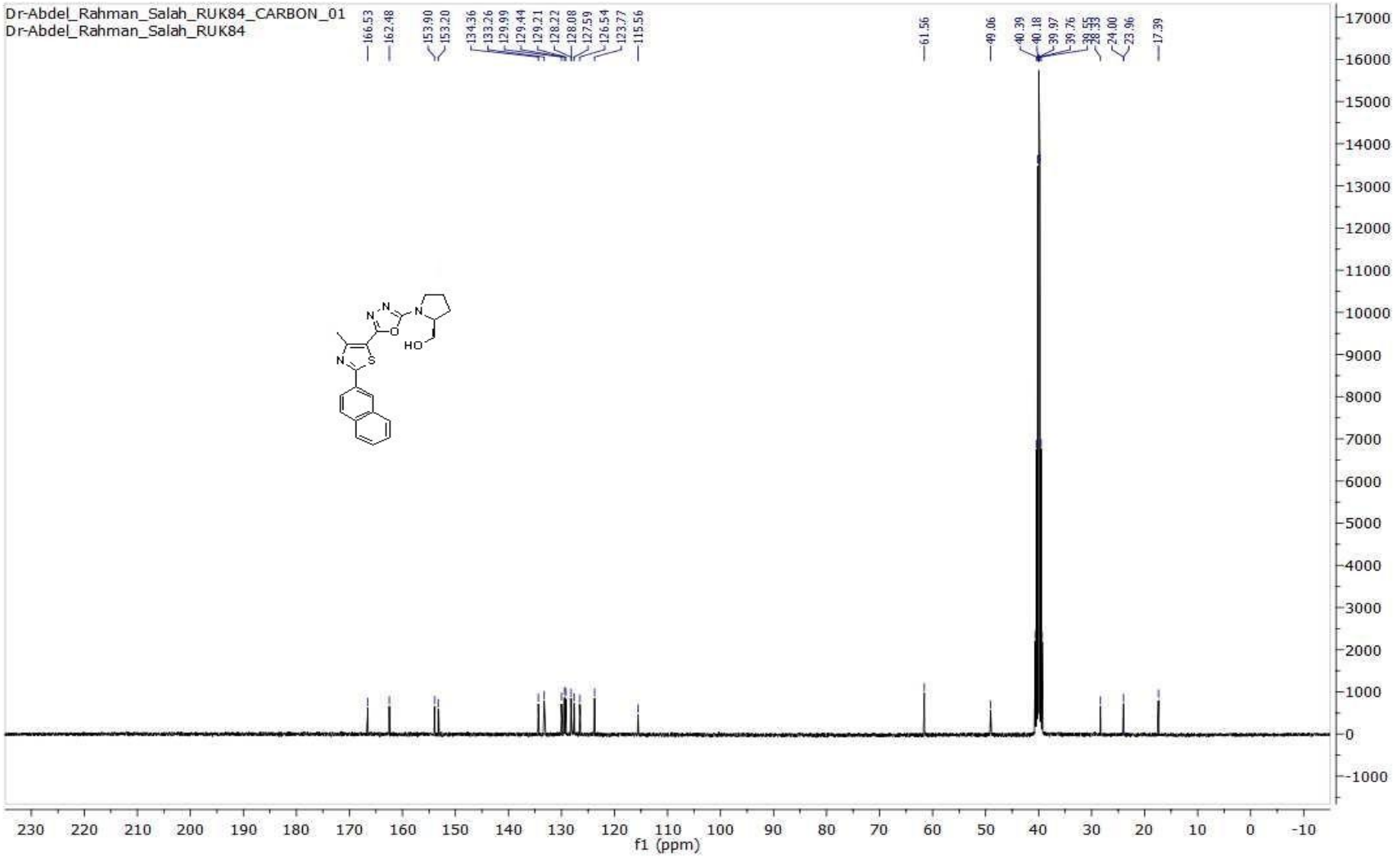
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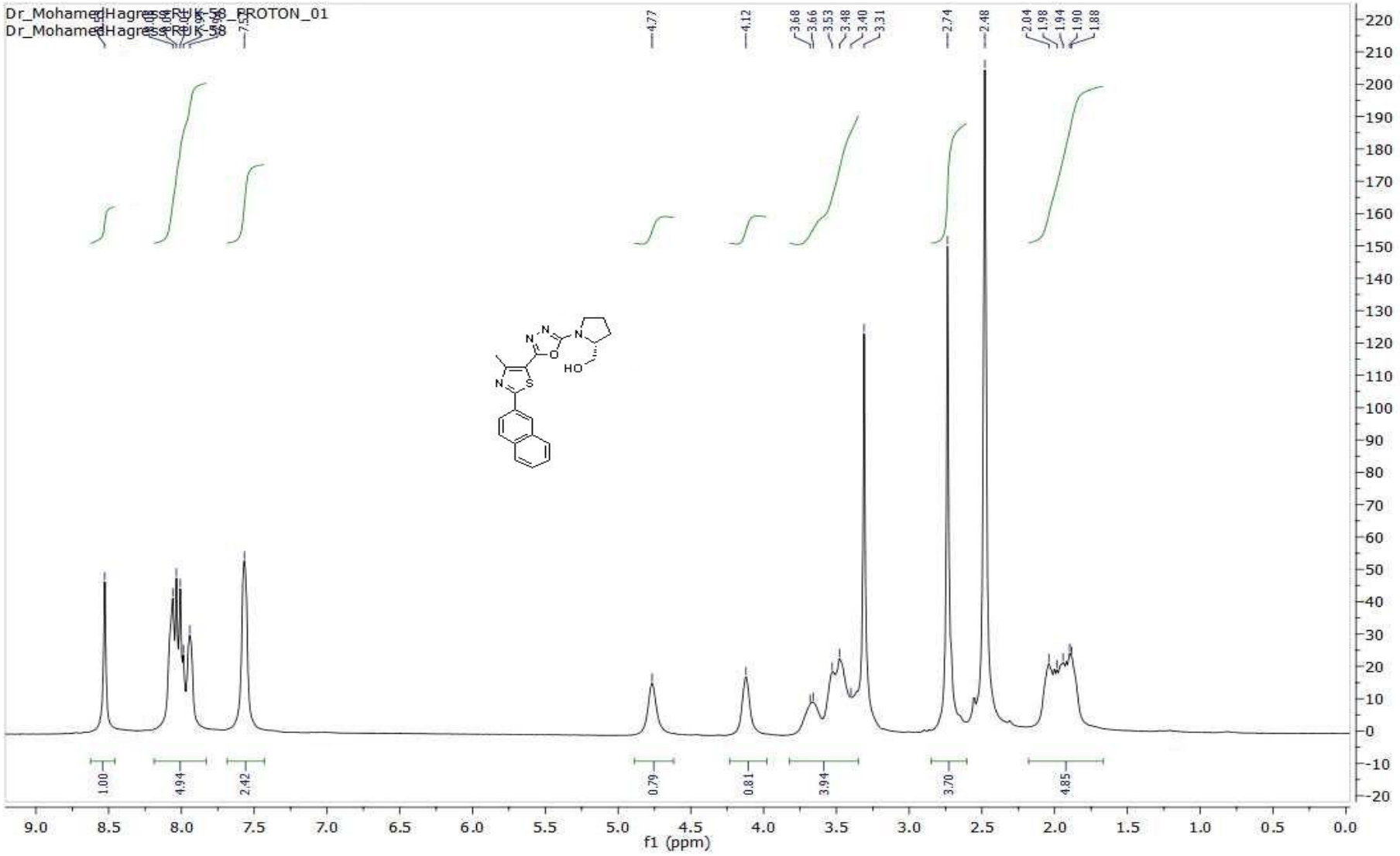
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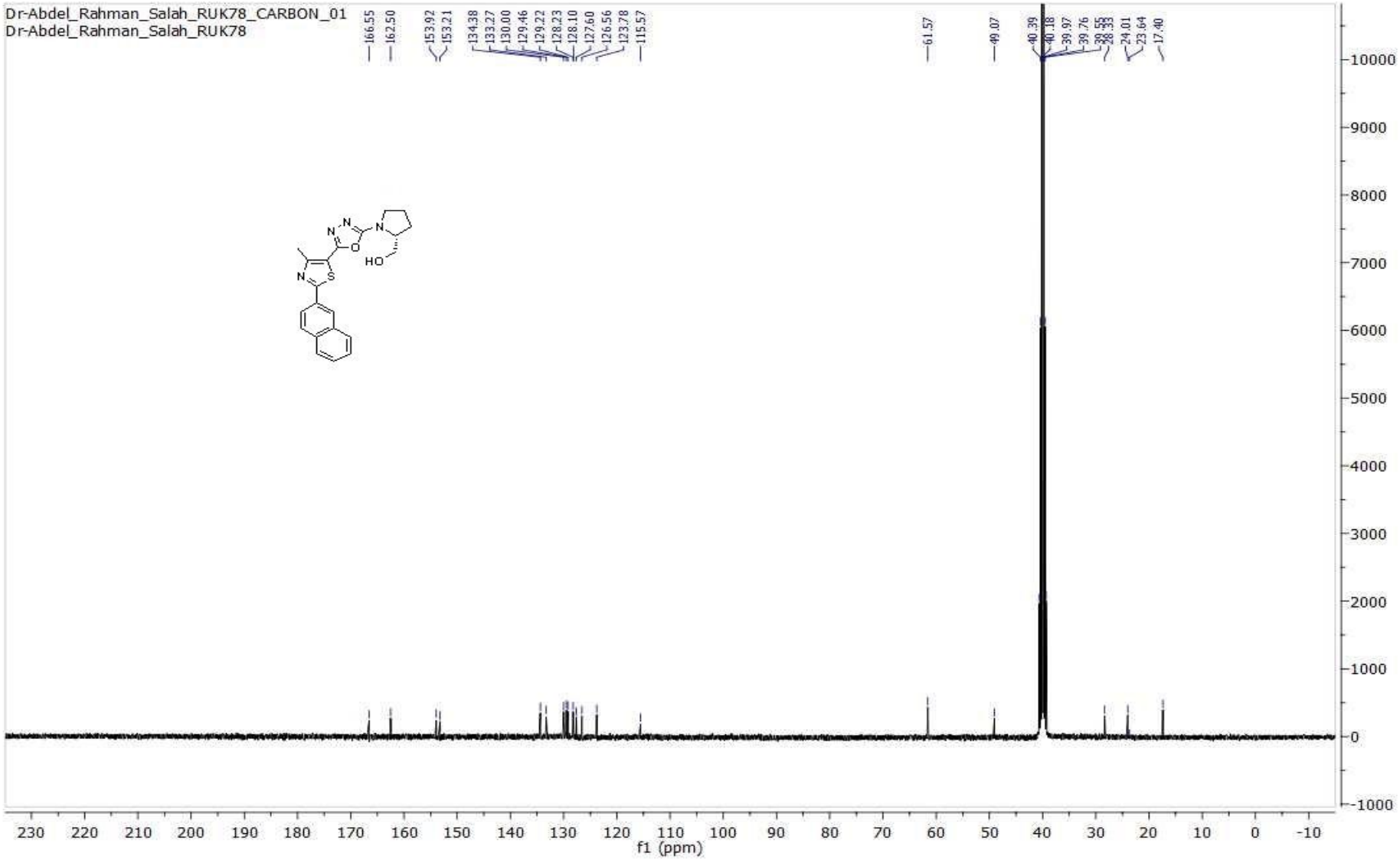
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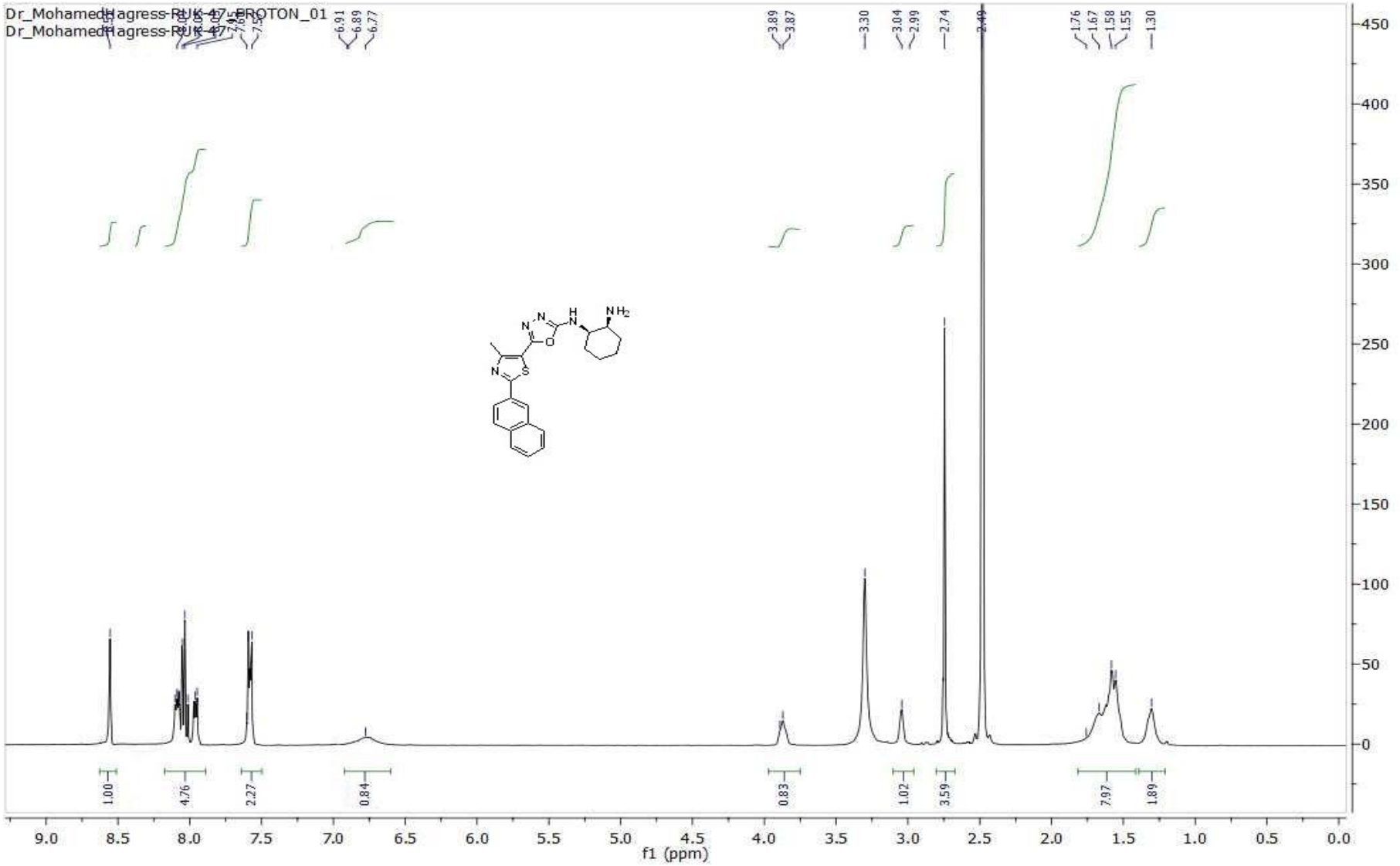
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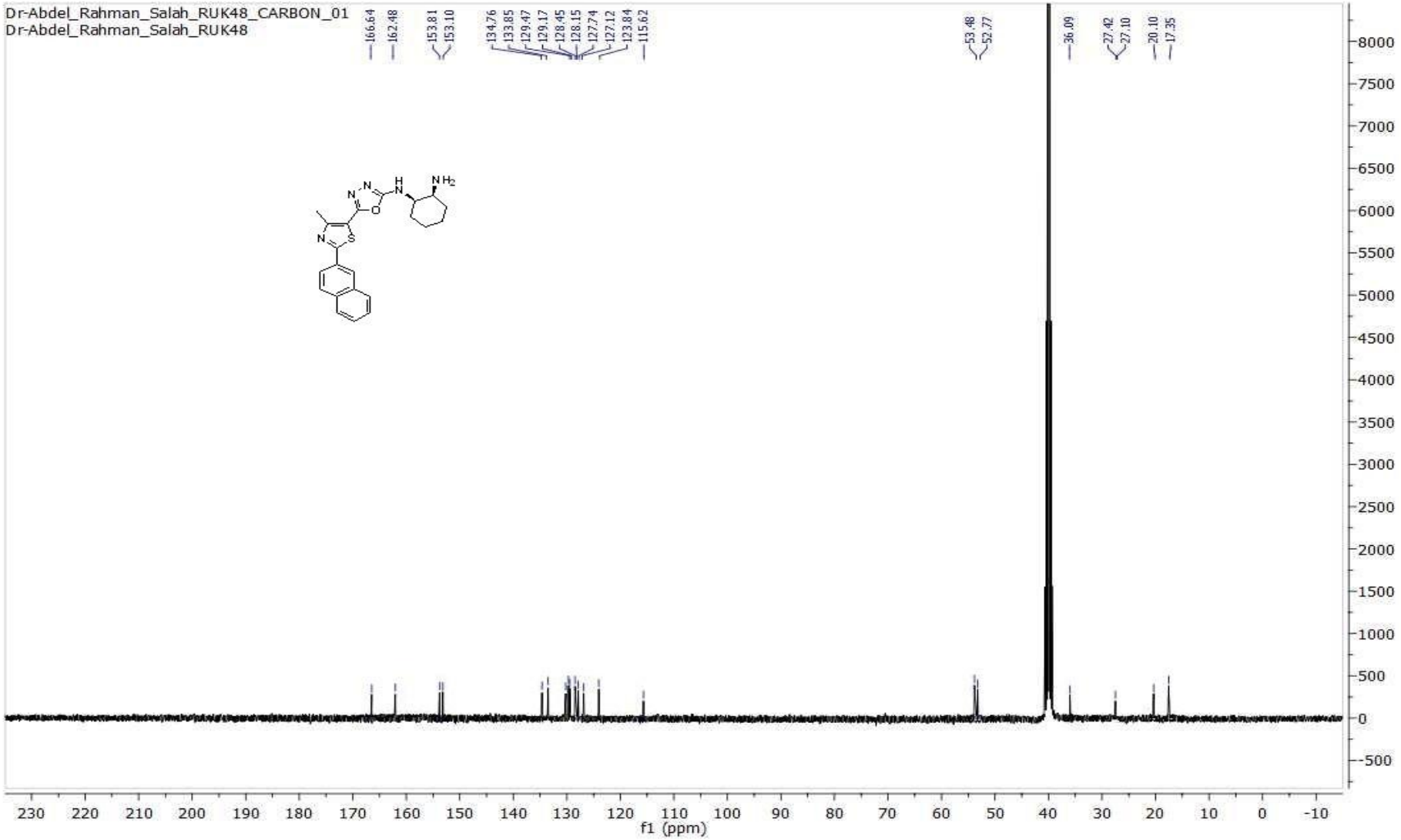
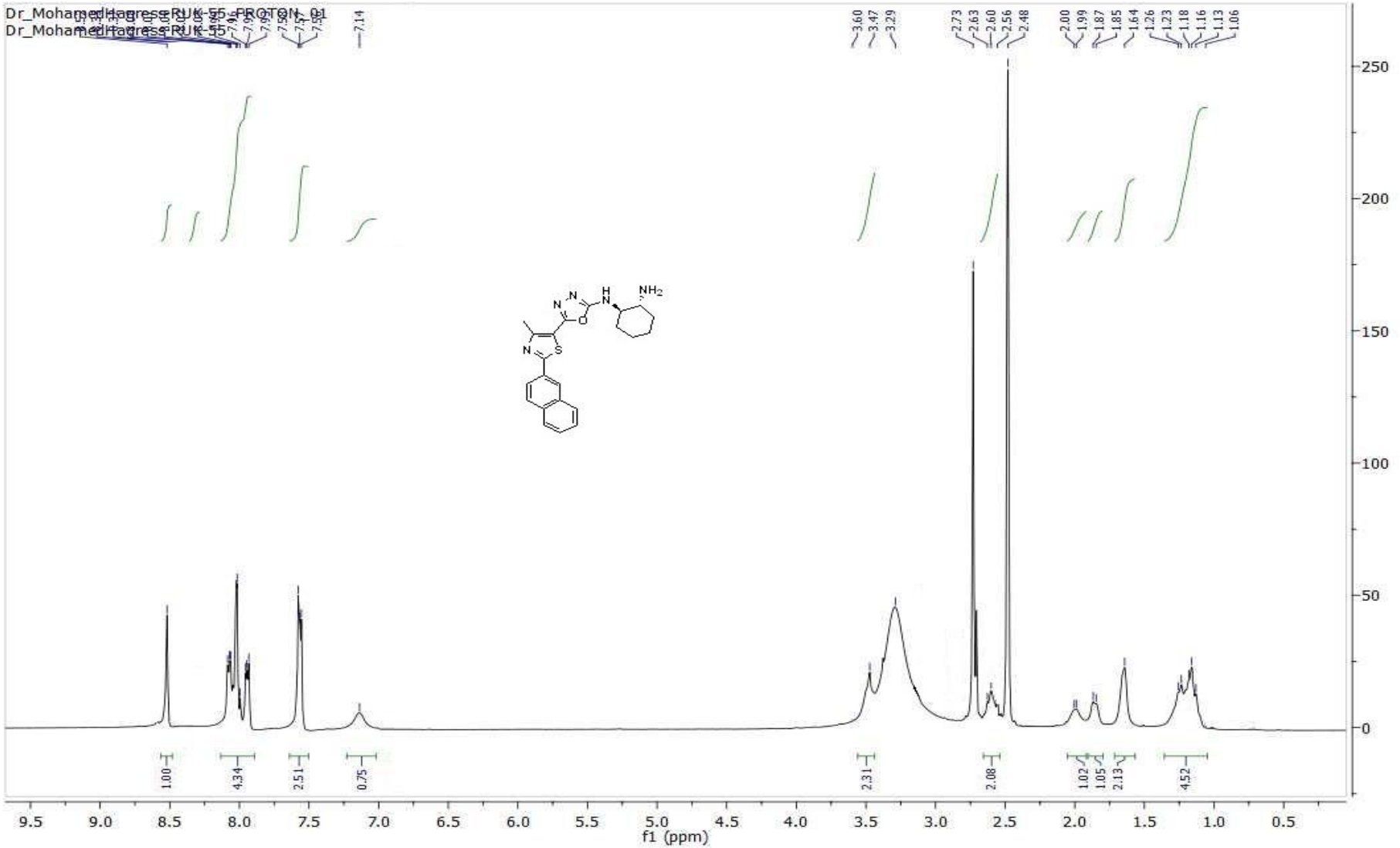
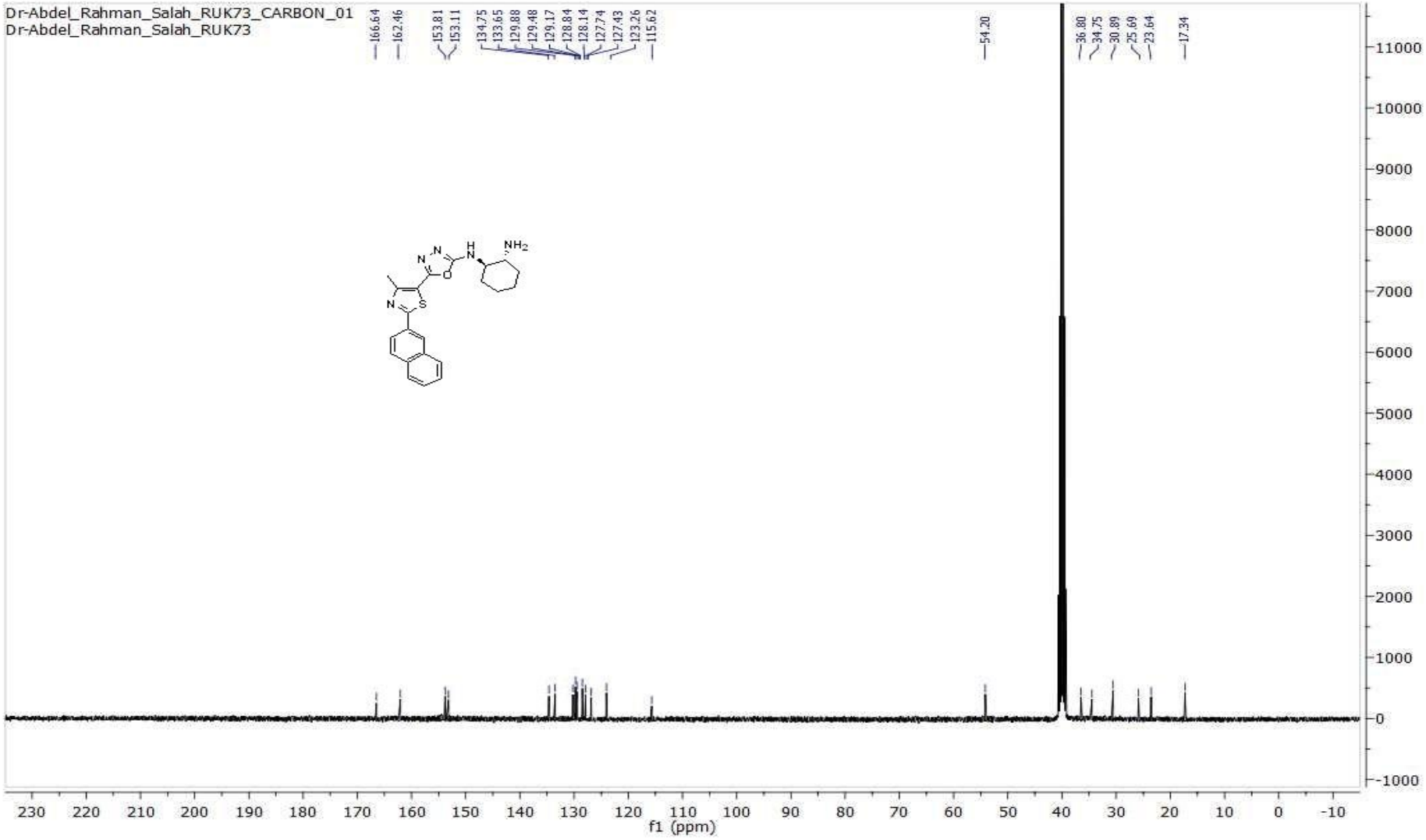
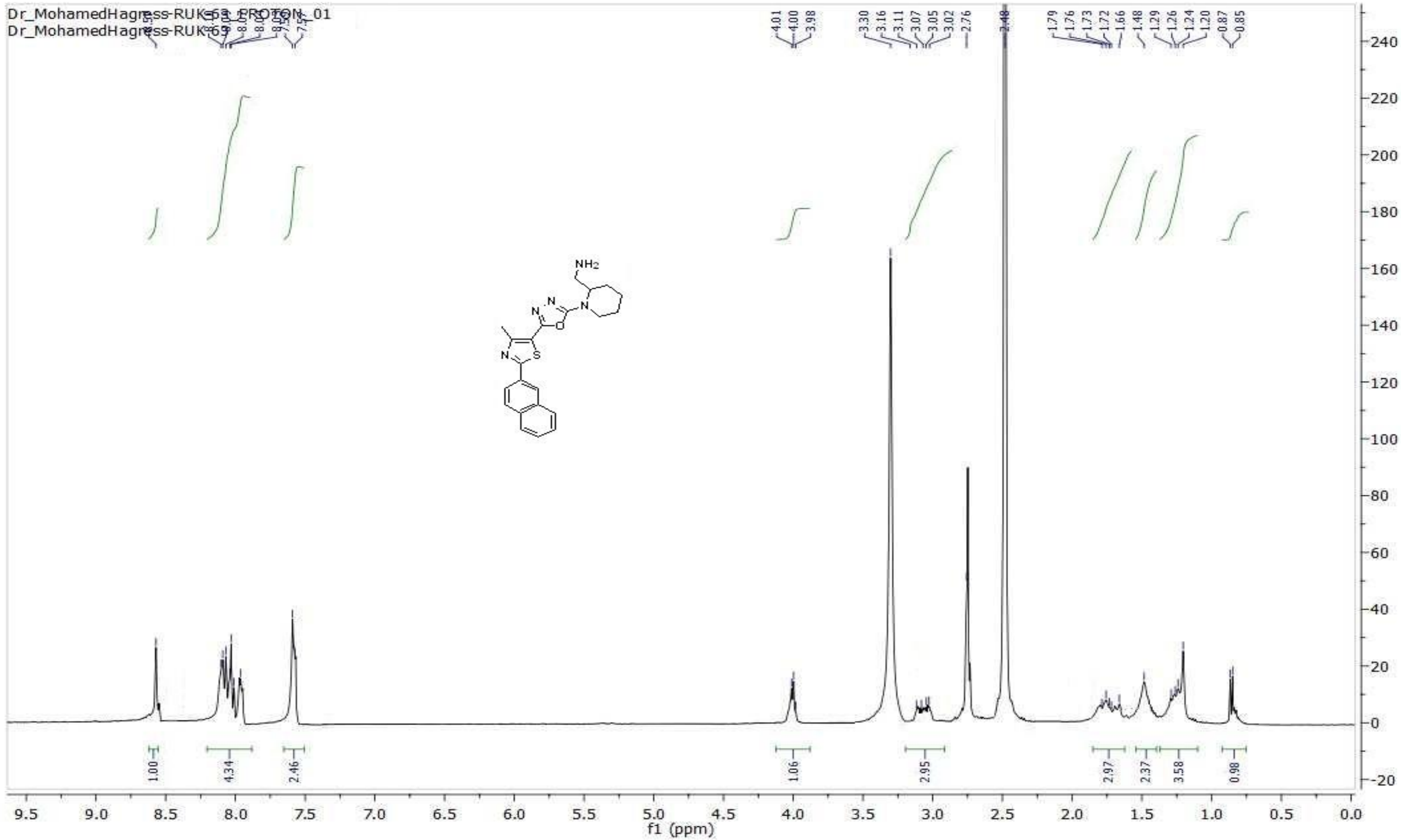
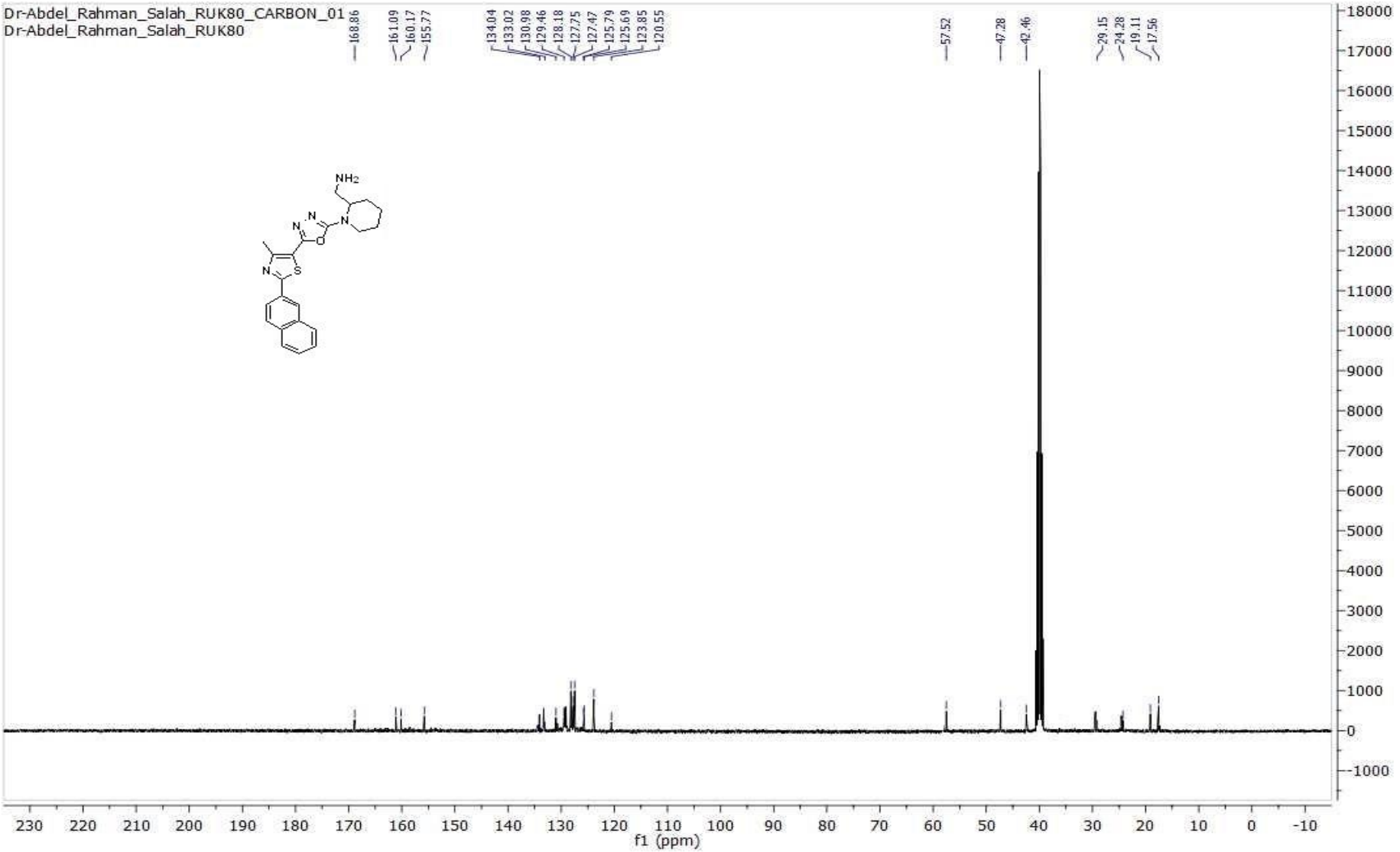
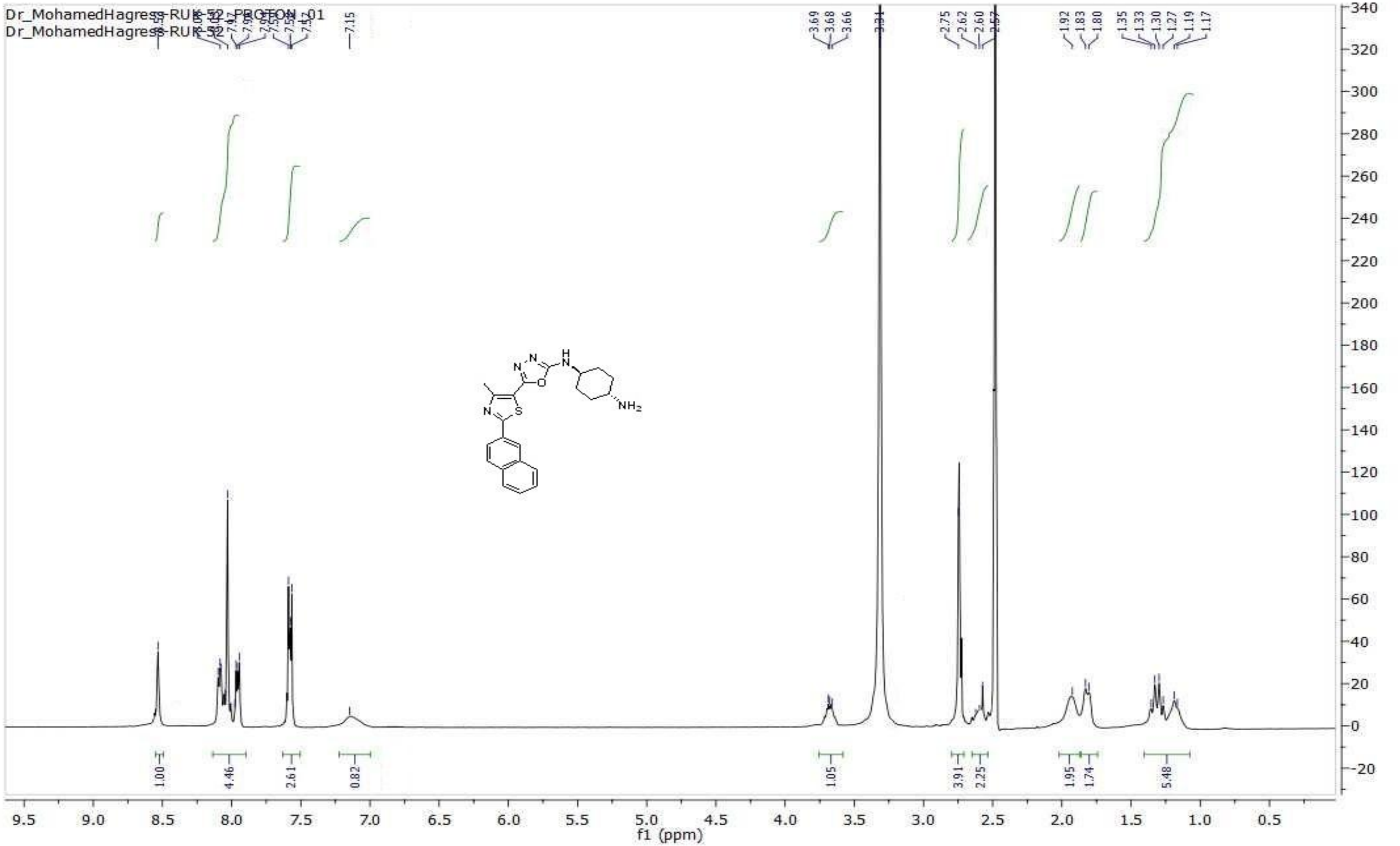

Dr-Abdel_Rahman_Salah_RUK48_CARBON_01
Dr-Abdel_Rahman_Salah_RUK48
166.64
162.48
153.81
153.10
134.76
133.85
129.47
129.17
128.45
128.15
127.74
127.12
123.84
115.62
53.48
52.77
36.09
27.42
27.10
20.10
17.35
NH2
f1 (ppm)

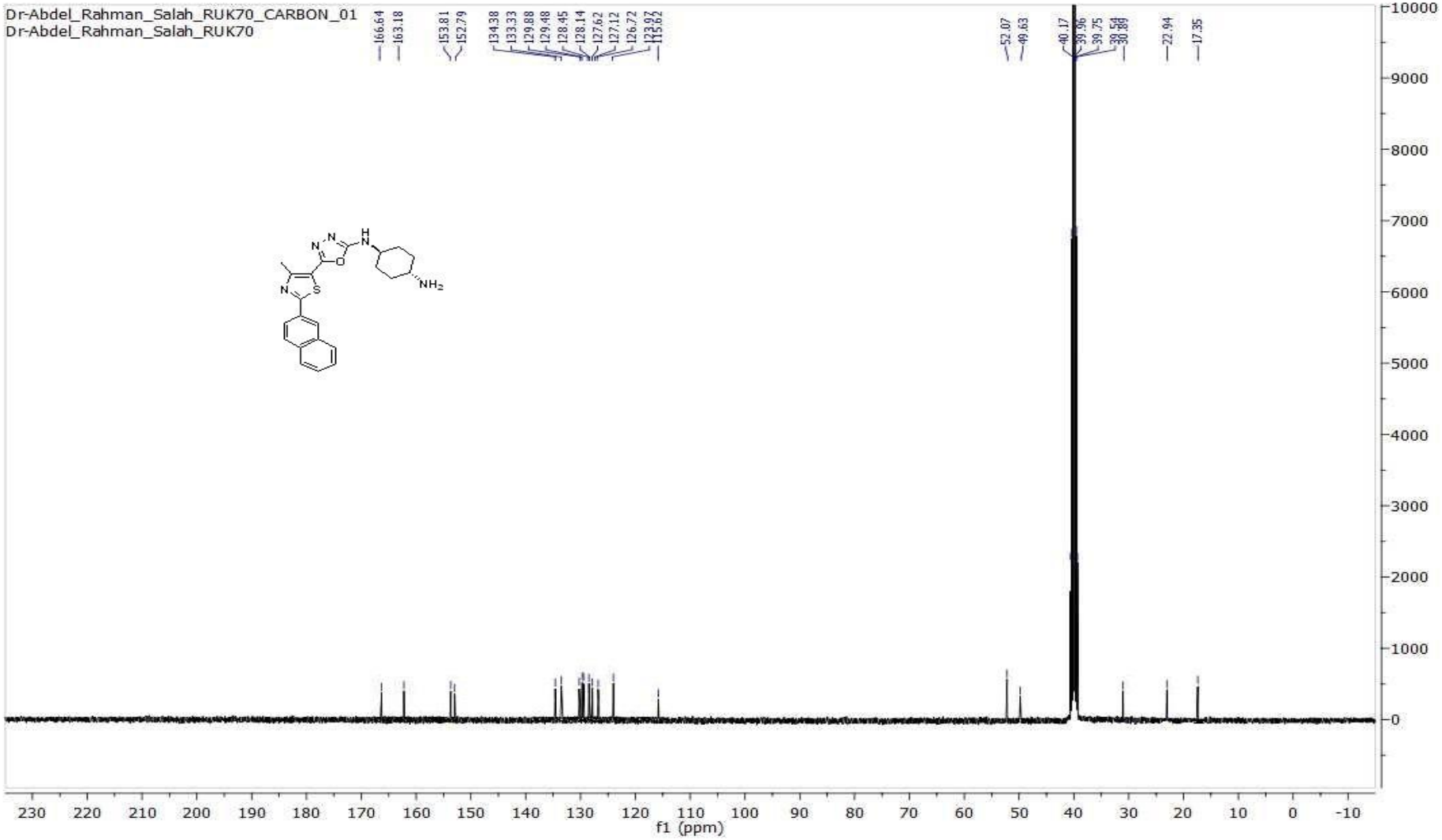

Dr_MohamedHagressRUK-55_PROTON_01
Dr_MohamedHagressRUK-55
7.14
3.60
3.47
3.29
2.73
2.63
2.60
2.56
2.48
2.00
1.99
1.87
1.85
1.64
1.26
1.23
1.18
1.16
1.13
1.06
NH2
1.00
4.34
2.51
0.75
2.31
2.08
1.02
1.05
2.13
4.52
f1 (ppm)

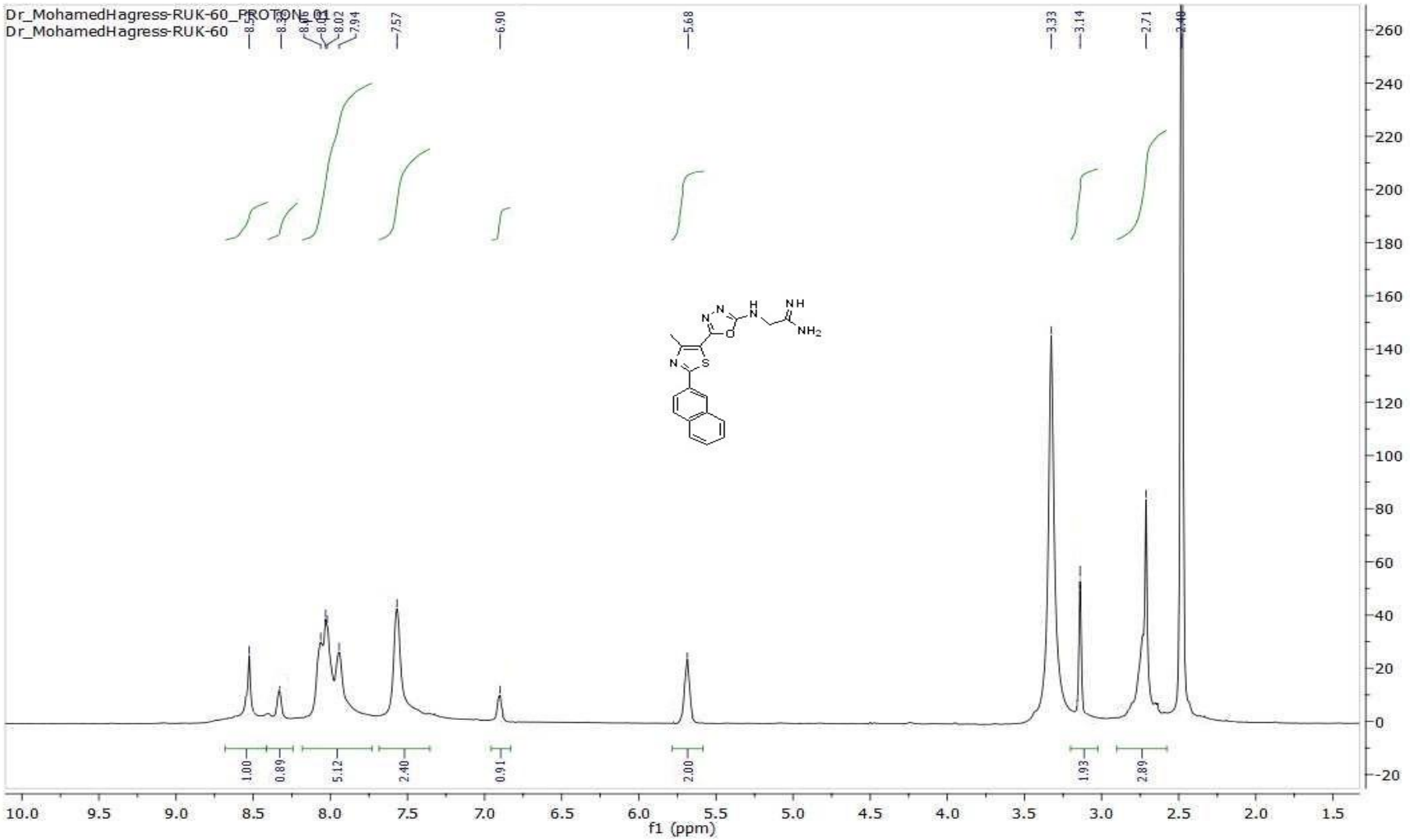

Dr-Abdel_Rahman_Salah_RUK73_CARBON_01
Dr-Abdel_Rahman_Salah_RUK73
166.64
162.46
153.81
153.11
134.75
133.65
129.88
129.48
129.17
128.84
128.14
127.74
127.43
123.26
115.62
54.20
36.80
34.75
30.89
25.69
23.64
17.34
11000
10000
9000
8000
7000
6000
5000
4000
3000
2000
1000
0
-1000
230 220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10
f1 (ppm)

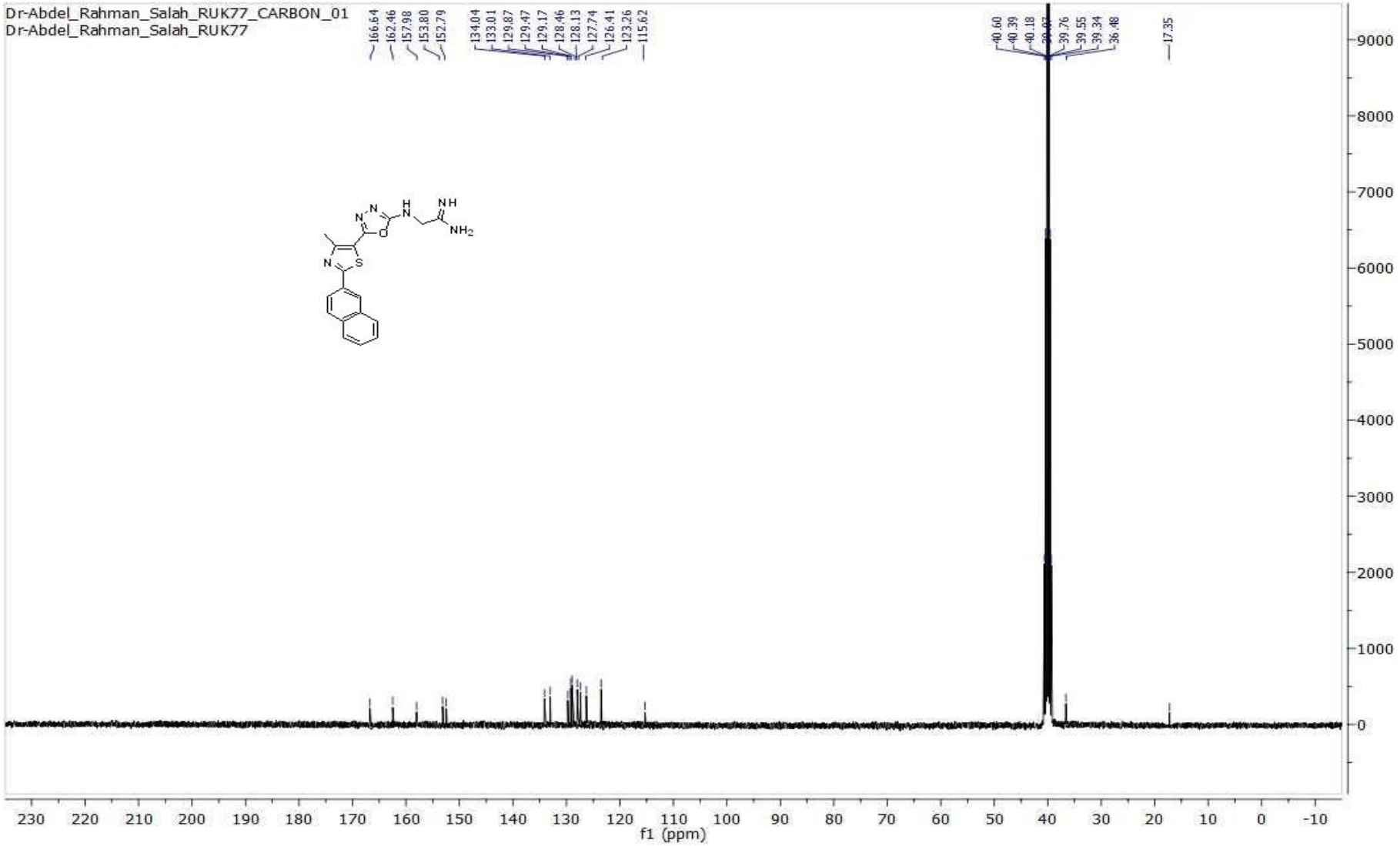

Dr_MohamedHagress-RUK-63_PROTON_01
Dr_MohamedHagress-RUK-63
4.01
4.00
3.98
3.30
3.16
3.11
3.07
3.05
3.02
2.76
2.48
1.79
1.76
1.73
1.72
1.66
1.48
1.29
1.26
1.24
1.20
0.87
0.85
NH2
N
O
S
1.00
4.34
2.46
1.06
2.95
2.97
2.37
3.58
0.98
f1 (ppm)


Dr-Abdel_Rahman_Salah_RUK80_CARBON_01
Dr-Abdel_Rahman_Salah_RUK80
168.86
161.09
160.17
155.77
134.04
133.02
130.98
129.46
128.18
127.75
127.47
125.79
125.69
123.85
120.55
57.52
47.28
42.46
29.15
24.28
19.11
17.56
NH2
N
N
O
N
S
18000
17000
16000
15000
14000
13000
12000
11000
10000
9000
8000
7000
6000
5000
4000
3000
2000
1000
0
-1000
230 220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10
f1 (ppm)


Dr_MohamedHagress-RUK-52_PROTON_01
Dr_MohamedHagress-RUK-52
7.15
3.69
3.68
3.66
3.31
2.75
2.62
2.60
2.57
1.92
1.83
1.80
1.35
1.33
1.30
1.27
1.19
1.17
1.00
4.46
2.61
0.82
1.05
3.91
2.25
1.95
1.74
5.48
f1 (ppm)
NH2


Dr-Abdel_Rahman_Salah_RUK70_CARBON_01
Dr-Abdel_Rahman_Salah_RUK70
166.64
163.18
153.81
152.79
134.38
133.33
129.88
129.48
128.45
128.14
127.62
127.12
126.72
115.82
52.07
49.63
40.17
39.96
39.75
22.94
17.35
NH2
f1 (ppm)


Dr_MohamedHagress-RUK-60_PROTON_01
Dr_MohamedHagress-RUK-60
8.52
8.39
8.14
8.03
8.02
7.94
7.57
6.90
5.68
3.33
3.14
2.71
2.49
1.00
0.89
5.12
2.40
0.91
2.00
1.93
2.89
f1 (ppm)


Dr-Abdel_Rahman_Salah_RUK77_CARBON_01
Dr-Abdel_Rahman_Salah_RUK77
166.64
162.46
157.98
153.80
152.79
134.04
133.01
129.87
129.47
129.17
128.46
128.13
127.74
126.41
123.26
115.62
40.60
40.39
40.18
39.97
39.76
39.55
39.34
36.48
17.35
NH
NH2
9000
8000
7000
6000
5000
4000
3000
2000
1000
0
230 220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10
f1 (ppm)